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TOWNSEND and TOWNSEND and CREW LLP

By: [signature]

PATENT
Attorney Docket No. 019904-002610US

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of:	Confirmation No. 5231
Joseph K. Belanoff	Examiner: Donna A. Jagoe
Application No.: 10/772,919	Technology Center/Art Unit: 1614
Filed: February 4, 2004	APPELLANTS' BRIEF UNDER 37 CFR §41.37
For: ANTIGLUCOCORTICOIDS FOR THE TREATMENT OF POSTPARTUM PSYCHOSIS	

Mail Stop Appeal Brief
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Commissioner:

Further to the Notice of Appeal mailed on August 19, 2009 for the above-referenced application, Appellants submit this Brief on Appeal.

A fee of $270 is required for a small entity filing an Appeal Brief, and the fee for a one-month extension of time, from October 19 to November 19, 2009 is $65. Please charge a total of $335 to Deposit Account Number 20-1430. No additional fees are believed to be necessary. If however, any additional fees are required, I authorize the Commissioner to charge these fees which may be required to Deposit Account Number 20-1430.

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1. REAL PARTY IN INTEREST

The real party in interest is Corcept Therapeutics, Inc. of Menlo Park, California.

2. RELATED APPEALS AND INTERFERENCES

This appeal has no related proceedings or interferences. Corcept owns US Application No. 11/519,008, directed to Methods of treating psychosis associated with interferon alpha therapy, that is also on appeal. The two applications are not related through priority.

3. STATUS OF CLAIMS

The claims in the application are 1-15.

Claims canceled: 12-14

Claims pending: 1-11, 15

Claims allowed: NONE

Claims rejected: 1-11, 15

Claims objected to: NONE

The claims on appeal are claims 1-11 and 15.

4. STATUS OF AMENDMENTS

An amendment was not filed after the May 19, 2009 Final Office Action. Claims 1-11 and 15 on appeal are as submitted in the Response to the Office Action filed on January 28, 2009.

5. SUMMARY OF CLAIMED SUBJECT MATTER

A. CLAIMS 1-11 and 15

Claims 1-11 and 15 are argued together for the issue of obviousness. Appellants argue that independent claim 1 is patentable and non-obviousness over the art, and thus, the subject matter of the dependent claims, which incorporate the limitations of claim 1, are also non-obvious. The subject matter of each claim is presented in turn below.

Claim 1 is directed to a method of ameliorating psychotic symptoms in a patient having postpartum psychosis (PPP) using a glucocorticoid receptor antagonist *(Specification, p. 1, line31, to p.2, line 4)*. The claim includes a proviso that the first psychotic symptoms arise within nine months of childbirth *(Specification, p. 4, lines 5-7)*, that the patient has never suffered any psychotic condition not triggered by childbirth, and that the patient did not suffer from psychosis prior to parturition (*i.e.*, childbirth) *(Specification, p. 4, lines 15-22)*.

Claim 2 further limits the time in which psychotic symptoms arise to eight weeks after childbirth *(Original claim 2)*.

Claim 3 specifies that the glucocorticoid receptor antagonist comprises a steroidal skeleton with at least one phenyl-containing moiety in the 11-β position of the steroidal skeleton *(Specification, p. 2, lines 5-7, p. 10, lines 17-31)*.

Claim 4 depends on claim 3, and further specifies that the phenyl-containing moiety in the 11-β position is a dimethylaminophenyl moiety *(Specification, p. 2, lines 7-8, p. 10, lines 17-31)*.

Claim 5 depends on claim 4, and further specifies that the glucocorticoid receptor antagonist is mifepristone *(Specification, p. 2, lines 8-9, p. 10, lines 17-18, Example 1)*.

Claim 6 also depends on claim 4, and further specifies that the glucocorticoid receptor antagonist is selected from the group consisting of 11β-(4-dimethylaminoethoxyphenyl)-17α-propynyl-17β-hydroxy-4,9 estradien-3-one and 17β-hydroxy-17α-19-(4-methylphenyl)androsta-4,9(11)-dien-3-one *(Specification, p. 2, lines 10-13, p. 10, lines 4-8)*.

Claim 7 depends on claim 1, and specifies that the glucocorticoid receptor antagonist is selected from the group consisting of 4α(S)-Benzyl-2(R)-prop-1-ynyl-1,2,3,4,4α,9,10,10a(R)-octahydro-phenanthrene-2,7-diol and 4α(S)-Benzyl-2(R)-chloroethynyl-1,2,3,4,4α,9,10,10a(R)-octahydro-phenanthrene-2,7-diol *(Specification, p. 2, lines 13-16, p. 12, lines 29-34).*

Claim 8 further specifies that the glucocorticoid receptor antagonist is (11β,17β)-11-(1,3-benzodioxol-5-yl)-17-hydroxy-17-(1-propynyl)estra-4,9-dien-3-one *(Specification, p. 2, lines 17-18, p. 12, line 34, to p. 13, line 2).*

Claim 9 further specifies that the glucocorticoid receptor antagonist is administered once per day *(Specification, p. 2, line 19, p. 22, lines 3-6).*

Claim 10 further specifies that the glucocorticoid receptor antagonist is administered orally *(Specification, p. 2, line 19-20, p. 16, line 16, to p. 18, line 19).*

Claim 11 further specifies that the glucocorticoid receptor antagonist is administered by transdermal application *(Specification, p. 15, lines 32-34, p. 18, line 30-32)*, nebulized suspension, or aerosol spray *(Specification, p. 2, line 20-21).*

Claim 15 further specifies that the glucocorticoid receptor antagonist is a specific glucocorticoid receptor antagonist *(Specification, p. 5, line 29, to p. 6, line 3).*

B. CLAIMS 3 AND 4 - DEPENDENT

Claims 3 and 4 are argued together for the issue of written description. Claim 3 depends on claim 1, and claim 4 depends on claim 3. The subject matter of these claims is set forth above.

6. GROUNDS OF REJECTION TO BE REVIEWED ON APPEAL

A. GROUND OF REJECTION 1 (Claims 1-11 and 15)

Claims 1-6 and 9-11 stand rejected under 35 USC § 103(a) as allegedly unpatentable over US Patent No. 6,150,349 to Schatzberg (hereinafter "Schatzberg ").

Claim 7 stands rejected under 35 USC § 103(a) as allegedly unpatentable over Schatzberg, in view of Stowe *et al.* (1995) *Am. J. Obstetrics Gynecol.* 173:639-45 (hereinafter "Stowe"), in view of Bradley *et al.* (2002) *J. Med. Chem.* 45:2417-24 (hereinafter "Bradley").

Claim 8 stands rejected under 35 USC § 103(a) as allegedly unpatentable over Schatzberg, in view of Stowe, in view of US Patent No. 6,011,025 to Gebhard (hereinafter "Gebhard").

Claim 15 stands rejected under 35 USC § 103(a) as allegedly unpatentable over Schatzberg, in view of Belanoff *et al.* (2002) *Curr. Psych. Reports* 4:164 (hereinafter "Belanoff").

As explained in Section 5 above, Appellants will argue the claims together for the issue of obviousness. Appellants consider independent claim 1 to be non-obvious over the art, and thus consider the dependent claims, which incorporate the limitations of claim 1, to also be non-obvious.

It is acknowledged that claims 1-6 and 9-11 are separately rejected under 35 USC § 103(a) as allegedly unpatentable over US Patent No. 6,362,173 to Schatzberg (hereinafter "Schatzberg II"). Similarly, claim 15 is separately rejected under 35 USC § 103(a) as allegedly unpatentable over Schatzberg II, in view of Belanoff. In making the separate rejections, the Examiner relies on passages from Schatzberg II that are identical to those recited in the rejection based on Schatzberg. Indeed, *Schatzberg II is a continuation of Schatzberg* and the specifications are identical. Both patents represent earlier work from the Appellants. Appellants therefore consider the arguments presented for the rejections based on Schatzberg to apply equally to the rejections based on Schatzberg II.

B. GROUND OF REJECTION 2 (Claims 3 and 4)

Claims 3 and 4 stand rejected under the first paragraph of 35 USC § 112 as allegedly lacking written description.

7. ARGUMENT

The following acronyms are used in this section:

GR:	glucocorticoid receptor
GRA:	glucocorticoid receptor antagonist
PPP:	postpartum psychosis
PMD:	psychotic major depression
DSM IV:	Diagnostic and Statistical Manual of Mental Disorders IV (2000)
IUPAC:	International Union of Pure and Applied Chemistry

A. GROUND OF REJECTION 1 (Claims 1-11 and 15)

Claims 1-6 and 9-11 stand rejected under 35 USC § 103(a) as allegedly unpatentable over Schatzberg. Claims 7, 8, and 15 are rejected as allegedly unpatentable over Schatzberg in view of various secondary references as discussed below. Appellants will argue the claims together for the issue of obviousness.

Claim 1 reads as follows:

> A method of ameliorating the psychotic symptoms of a patient having postpartum psychosis, comprising administering an amount of a glucocorticoid receptor antagonist effective to ameliorate the psychotic symptoms of the postpartum psychosis, with the proviso that the first psychotic symptoms arise within nine months of childbirth, that the patient has never suffered any psychotic condition not triggered by childbirth, and that the patient did not suffer from psychosis prior to parturition.

In summary, this appeal presents the question of which party bears the burden of providing evidence. The prior art discloses that some, but not all, psychotic conditions are due to glucocorticoid dysregulation, and thus amenable to treatment with a GRA. The same prior art includes PPP among the psychotic conditions known to afflict human beings. The Examiner places upon the Appellants the heavy burden of proving a negative. That is, the Examiner repeatedly asserts that, absent evidence from the prior art that a given psychotic condition is **NOT** intended to be included among those amenable to treatment with a GRA, it will be viewed

as such. Appellants have submitted rebuttal evidence teaching away from treating a postpartum mother with a GRA, because glucocorticoid levels fall dramatically after birth. Applicants urge that the Patent Office has not established that the prior art suggests that PPP is caused by glucocorticoid dysregulation.

Rejection

The first Office Action issued by the present Examiner on September 30, 2008, includes a summary of Schatzberg. According to the Examiner, Schatzberg teaches GRAs, including those recited in claims 3-6 on appeal, for treatment of psychosis. The Examiner states that Schatzberg teaches that conditions involving psychosis can include psychotic disorders not otherwise specified. The Examiner points to the disclosure from the DSM IV, which includes PPP as an example of a psychotic disorder not otherwise specified. The Examiner also states that Schatzberg teaches daily administration of GRAs both orally and transdermally, as recited in claims 9-11 on appeal (September 30, 2008 Office Action, p.6).

The Examiner concludes that it would have been obvious to employ the recited GRAs for amelioration of the symptoms of PPP motivated by the teaching of Schatzberg I who teach that GRAs ameliorate some types of psychosis (September 30, 2008 Office Action, p.6-7).

A rejection based on Schatzberg had already been overcome with the previous Examiner once the disclosure of Schatzberg was explained in more detail, and distinguished from the present invention (*see* June 18, 2008 Interview Summary and July 11, 2008 Response). Appellants therefore scheduled an interview with the present Examiner to more efficiently dispatch with the issue.

During the Interview of January 7, 2009, Appellant's representative explained that Schatzberg discloses treatment of psychotic symptoms associated with glucocorticoid regulatory dysfunction, not psychosis generally. PPP is included in a section of Schatzberg that reviews psychosis generally, and includes an exhaustive list of conditions. Appellant's representative explained that Schatzberg expressly excludes psychotic conditions that are not associated with glucocorticoid regulatory dysfunction. For example, Schatzberg states that the psychotic symptoms of schizophrenia as not included in the invention.

The Examiner stated that, unless the condition was ***explicitly excluded*** from the invention of Schatzberg, that it would be obvious for one of skill to treat the psychotic symptoms with a GRA as taught by Schatzberg (*see* January 12, 2009 Interview Summary).

The Office Action issued May 19, 2009 includes a response to Appellant arguments on pages 16-17. The Examiner asserts that Schatzberg teaches treatment of psychosis with a GRA, and that the psychotic conditions listed in the reference are "not for informational purposes." According to the Examiner, the psychotic conditions listed in Schatzberg "are the list of those treatable with a GRA." The Examiner points to the disclosure of PPP in Schatzberg, and repeats the assertion that, because the reference does not ***specifically exclude*** PPP from treatment with a GRA, that it is included.

The Examiner does not accept the Appellant argument that the Examiner has not provided evidence linking PPP with glucocorticoid regulatory dysfunction. According to the Examiner, this evidence is supplied by Schatzberg. The Examiner also does not acknowledge the evidence submitted by the Appellants that teaches away from treating a mother with a GRA soon after giving birth.

Finally, on page 17, the Examiner states that Appellant remarks disparage Schatzberg, implying that Appellants question the validity of the claims in Schatzberg.

Legal standard for obviousness

The Supreme Court in *KSR* warned against overly rigid application of the so-called teaching/ suggestion/ motivation (TSM) test of obviousness and essentially expanded the field of knowledge from which a motivation to modify or combine the references could be drawn. The motivation to combine may be explicit or implicit and may be found in the knowledge of one of ordinary skill in the art, scientific principles, or legal precedent (*see* MPEP § 2144).

A *prima facie* case of obviousness still requires the Examiner to establish that one of skill in the art would have a reasonable expectation of success in making the claimed invention (*see* MPEP § 2143.02). *KSR* reaffirmed the importance of predictability in

determining obviousness (*see* MPEP 2141, where the word "predictable" features prominently among potential rationales for a *prima facie* case).

Predictability is based on the person of ordinary skill **at the time of the invention**. Indeed, the MPEP 2142 explains that the examiner must step backward in time and into the shoes worn by the hypothetical person of ordinary skill in the art when the invention was unknown.

MPEP 2142 states that rejections for obviousness cannot be sustained with mere conclusory statements. The burden is on the examiner, in view of all factual information, to provide some articulated reasoning with some rational underpinning to support the legal conclusion of obviousness. The examiner must provide evidence which **as a whole** shows that the legal determination sought to be proved is more probable than not.

The legal concept of *prima facie* obviousness allocates who has the burden of going forward with production of evidence in each step of the examination process. **The examiner bears the initial burden** of factually supporting any *prima facie* conclusion of obviousness. If the examiner does not produce a *prima facie* case, **the applicant is under no obligation to submit evidence of nonobviousness**. If, however, the examiner does produce a *prima facie* case, the burden of coming forward with evidence or arguments shifts to the applicant, who may submit additional evidence of nonobviousness (MPEP 2142). MPEP 2141.02 again emphasizes that **prior art must be considered in its entirety**, including disclosures that teach away from the claims.

Schatzberg does not teach that all psychotic symptoms are treatable with a GRA, only those associated with glucocorticoid regulatory dysfunction

Appellants have repeatedly explained that Schatzberg limits the scope of their invention to treatment of psychotic conditions that are associated with glucocorticoid regulatory dysfunction. The focus of the patent is on psychotic major depression, and the claims are limited to amelioration of psychosis associated with major depression. Appellants note the following passages from Schatzberg:

INTRODUCTION

This invention is directed to a method for treating psychosis whose pathogenesis is related to glucocorticoid regulatory dysfunction.

...

SUMMARY OF THE INVENTION

The invention is directed to a method of treating psychosis associated with glucocorticoid related dysfunction by administration of an amount of a glucocorticoid receptor antagonist effective to ameliorate the psychosis, with the proviso that the patient not be suffering from Cushing's Syndrome.

Schatzberg makes clear that not all psychotic symptoms can be treated with a GRA. The disclosure in Col. 6 explains that psychotic symptoms in schizophrenic and manic patients are not related to the glucocorticoid pathway.

Most psychotic patients have a glucocorticoid regulatory dysfunction (as indicated by non-responsiveness in the DS test). In contrast, patients with, e.g., schizophrenia (including those historically described as "psychotic schizophrenics") and manic states, do not have glucocorticoid regulatory dysfunction (as indicated by responsiveness in the DS test).

While PPP is included in a general disclosure of conditions that give rise to psychotic symptoms, nowhere does Schatzberg state that the psychotic aspect of PPP results from glucocorticoid dysregulation.

Additional teachings from Schatzberg are set forth in more detail in the following sections.

The claimed methods are distinguishable from the teaching of Schatzberg

Application of the law to the facts in this case is a straightforward exercise. Schatzberg teaches that GRAs can be used to treat psychotic conditions associated with glucocorticoid regulatory disfunction. Schatzberg teaches that **not all** psychotic conditions **ARE** associated with glucocorticoid regulatory dysfunction, and specifically points to the psychotic

symptoms of schizophrenic patients as not associated with glucocorticoid signaling. A fair reading would not lead a reader to conclude that Schatzberg teaches the use of GRAs for treating any and all forms of psychosis.

In order for the present claims to be obvious, one of skill, at the time of the present invention, would have to have a reasonable expectation of success in treating PPP with a GRA. That means that there must be some teaching, suggestion, or knowledge common among those in the art that PPP is associated with glucocorticoid regulatory dysfunction.

The Examiner has not provided such evidence, as explained in more detail in section A.5 below. One of skill would therefore not have a reasonable expectation that a GRA could be used to treat every psychotic syndrome, or PPP in particular. The Exhibits described below demonstrate the difficulty associated with distinguishing the underlying causes of any particular psychiatric symptom. Syndromes that entail similar outward symptoms may have entirely distinct etiologies. The Exhibits relating to PPP confirm that this particular phenomenon was not well understood.

In the Response submitted October 24, 2007, Appellants submitted a Declaration under 37 CFR 1.132 from Dr. Joseph Belanoff. In his Declaration, Dr. Belanoff presented results showing that, while the GRA mifepristone effectively reduced psychotic symptoms of patients with psychotic major depression, it was not effective in patients with schizoaffective disorders. The Belanoff Declaration is included in the Evidence Appendix as **Ex. A**.

The Declaration and associated evidence demonstrate that GRAs are not effective for treating psychosis generally. The etiology of any given psychiatric condition can be complicated and multifactorial; similar symptoms are not always effectively ameliorated in the same way.

Schatzberg claims the use of GRAs for treating the psychotic symptoms of psychotic major depression (PMD). Schatzberg also includes a general definition of psychosis from the DSM IV. An excerpt from the DSM IV was included in the Response submitted February 13, 2008 (listed in the Evidence Appendix as **Ex. B**). The DSM IV authors provide a subclassification of various psychotic disorders that are not well understood and do not meet

other DSM IV categories. Among these psychotic disorders is PPP. There is no suggestion that PPP is associated with glucocorticoid dysregulation.

The next set of Exhibits was submitted with the February 13, 2008 Response to distinguish postpartum psychosis from other postpartum psychiatric disorders.

Kaplan and Sadocks Comprehensive Textbook of Psychiatry (2000) has a chapter on postpartum psychiatric disorders (**Ex. C**). On page 1278, the authors identify three distinct postpartum disorders: blues, depression, and psychosis. The text explains that PPP is the most severe and rare postpartum psychiatric disorder. The last lines reveal the debate in the medical community as to how to characterize PPP, indicating that it was not well understood as of 2000. **Ex. D** is an article from the Peace and Healing website that explains that PPP is not a variant of postpartum depression, but a distinct entity. Finally, **Ex. E** is an article from National Public Radio, recounting one mother's experience with PPP. She explains that her psychotic episode arose without warning or depression. The article states that psychiatrists weren't sure (as of 2002) of what causes the syndrome.

Nothing in any of these articles suggests that PPP was known to be associated with glucocorticoid regulatory dysfunction. If anything, the articles emphasize the rare and mysterious nature of the disorder. Taken as a whole, the references indicate that the cause of PPP was not understood, and demonstrate the art-recognized difficulty in finding the distinct causes of what appear to be similar psychiatric symptoms.

The Examiner has not applied the proper standard for obviousness

As explained above, in order for the present claims to be obvious, the Examiner must provide reasons why one of skill, at the time of the present invention, would have to have a reasonable expectation of success in treating PPP with a GRA. Given that Schatzberg teaches GRA therapy for psychotic symptoms associated with glucocorticoid regulatory dysfunction, the Examiner must provide evidence of some teaching, suggestion, or knowledge common among those in the art at the time that PPP is associated with glucocorticoid regulatory dysfunction.

The Examiner has not provided such evidence. Appellant is unaware of evidence, documentary or otherwise, suggesting that one of skill at the time of the invention would reasonably expect the psychotic symptoms PPP to result from glucocorticoid dysregulation.

The Examiner points to the section in Schatzberg that discusses and defines psychotic conditions generally. According to the Examiner, unless the disclosure of Schatzberg ***specifically excludes*** a particular psychotic condition from the invention, one of skill would expect the condition to be included in the invention (*see* January 12, 2009 Interview Summary and May 19, 2009 Office Action).

The rationale that an invention is obvious absent a teaching that specifically excludes or teaches away from the invention does not reflect the law. Instead, a showing of obviousness requires an Examiner to provide articulated reasoning with some rational underpinning for making the legal conclusion, based on the evidence **as a whole**.

Essentially, the Examiner has shifted the burden to the Appellants to provide evidence teaching away from the present invention. This before meeting the burden of making a *prima facie* case by establishing a connection between PPP and cortisol dysregulation.

The Examiner has mischaracterized the teaching of Schatzberg

Schatzberg discloses that psychosis associated with glucocorticoid regulatory dysfunction can be treated with a GRA. The focus of the application is on psychotic major depression, and the claims are limited to methods of treating psychosis associated with major depression.

The Examiner states that Schatzberg teaches treatment of psychosis with a GRA. The Examiner asserts that Schatzberg teaches that the list of psychotic conditions included in the patent are treatable with a GRA. The Examiner states that PPP is included in this list, and therefore treatable with a GRA as taught by Schatzberg. According to the Examiner, schizophrenia is specifically excluded from the invention.

These assertions mischaracterize the teaching of Schatzberg, and does not appreciate its teachings as a whole. Appellants again point to the first sentences of the Introduction and Summary of the Invention, which specify that the invention is directed to

treating psychosis *associated with glucocorticoid regulatory dysfunction* (*see* Section A.3 above). The Examiner's characterization of the reference ignores the "related to/ associated with glucocorticoid regulatory dysfunction" clause.

PPP is disclosed in a section that describes psychotic conditions generally. In Col. 5, Schatzberg states that, "[t]he term 'psychotic' as used herein refers to a psychiatric condition in its broadest sense, as defined in the DSM-IV ... and described below." Schatzberg does not limit the term "psychotic" to those conditions related to glucocorticoid regulatory dysfunction – it is a general term.

The section that describes psychotic conditions and diagnosis (beginning in Col. 12 of Schatzberg) recites the following broad introduction, and continues for several pages, describing dozens of conditions that include psychotic components.

> 3. DIAGNOSISNG AND ASSESSING CONDITIONS AND ILLNESSES INVOLVING PSYCHOSIS
>
> Psychosis can be manifested as a mental illness in the form of a syndrome or as an element of a variety of disease processes. There are various means to diagnose these various forms of psychosis and assess the success of treatment.

PPP is included in Col. 15 as a "psychotic disorder not otherwise specified." Notably, **in Col. 16 of the same section, Schatzberg describes schizophrenia**. Thus, while Col. 6 explains that psychosis related to schizophrenia and manic states does not result from glucocorticoid regulatory dysfunction, the condition is still described. Contrary to the assertion by the Examiner, the disclosure of PPP in this general section does not necessarily indicate that it is treatable with a GRA.

Moreover, on page 17 of the May 19, 2009 Office Action, the Examiner asserts that the Appellants have disparaged Schatzberg, and that a patent shall be presumed valid. Appellant's remarks in no way disparage this patent. The inventor of the present application is a co-inventor of Schatzberg. Appellants note that the claims of Schatzberg are limited to methods of ameliorating psychosis associated with major depression, which is described in the

specification as associated with glucocorticoid regulatory dysfunction. Appellants have never asserted that the claims of Schatzberg encompass treatment of all psychotic conditions.

Importantly, Schatzberg never associates PPP with glucocorticoid regulatory dysfunction. As explained above, Appellants are unaware of evidence from the time of the invention showing association of PPP with the glucocorticoid pathway, and it has not been provided by the Examiner.

In summary, Schatzberg teaches that psychosis ***associated with glucocorticoid regulatory dysfunction*** can be treated with a GRA. Schatzberg does not teach that PPP is associated with glucocorticoid regulatory dysfunction. The mere disclosure of PPP in a general section describing conditions with psychotic components does not imply that the condition is treatable with a GRA, as evidenced by the fact that schizophrenia is also included in this section.

The Examiner has not acknowledged evidence teaching away from the present claims

Making a determination of obviousness requires consideration of the record as a whole. While not recognizing the existence of a *prima facie* case of obviousness, Applicants submitted rebuttal evidence teaching away from treating a postpartum mother with a GRA with the response dated January 28, 2009. Although properly entered, this evidence was not acknowledged by the Examiner in the May 19, 2009 Office Action.

The prior art teaches that cortisol levels fall dramatically immediately after birth. The presently claimed methods involve inhibition of cortisol signaling, *i.e.*, using a GRA. One of skill would not be motivated to treat a woman immediately postpartum with an antagonist of a hormone that is already in rapid decline.

The first page of Elenkov *et al.* explains that cortisol, as well as other hormones, increases in late pregnancy and falls rapidly in the early postpartum period (**Ex. F**, page 4933, last paragraph). Hendrick *et al.* (**Ex. G**) dedicate an entire section to cortisol in the postpartum period. The report confirms that cortisol levels fall abruptly at delivery (page 97, col. 2).

The art thus indicates that cortisol levels fall dramatically after birth. Absent some evidence that glucocorticoid regulatory dysfunction was linked with the psychotic

symptoms of PPP, one of skill would not be motivated to treat a postpartum mother with an inhibitor of cortisol signaling. As explained above, the Examiner has not provided this evidence.

Instead, the Examiner again cited Schatzberg, as on page 16 of the May 19, 2009 Office Action:

> Applicant repeatedly states that the Examiner has not provided evidence from the relevant time period to support a medically accepted link between glucocorticoid regulatory dysfunction and PPP. In response, the Examiner has provided the Schatzberg patent.

No mention was made of the rebuttal evidence from the Appellants. The Examiner has not met the burden of providing evidence in support of a *prima facie* case of obviousness OR of responding to rebuttal evidence from the Appellants.

The prior art discloses (1) that psychotic conditions associated with glucocorticoid dysregulation can be treated with a GRA and (2) cortisol levels fall dramatically in the mother after giving birth. One of skill, considering the knowledge in the art at the time, would not be motivated to treat a mother with the rare disorder of postpartum psychosis with a GRA.

The secondary references do not cure the deficiencies in the Examiner's arguments based on Schatzberg

Claim 7 stands rejected under 35 USC § 103(a) as allegedly unpatentable over Schatzberg, in view of Stowe, in further view of Bradley. Claim 8 is similarly rejected over Schatzberg, in view of Stowe, in view of Gebhard.

It is unclear why the Examiner has cited Stowe for these rejections, and not the primary rejection. Stowe focuses on postpartum depression, not PPP. In any event, Stowe does not teach or suggest that PPP is related to glucocorticoid regulatory dysfunction, and thus does not cure the defects of the Examiner's rejection based on Schatzberg.

According to the Examiner, Bradley teaches the GRA compounds recited in claim 7 (*see* page 12 of the September 30, 2008 Office Action). The Examiner concludes that one of skill would recognize the ability to substitute compounds that have the same GRA properties and have an obvious reasonable expectation of success.

According to the Examiner, Gebhard teaches the GRA compounds recited in claim 8 (*see* page 12-13 of the September 30, 2008 Office Action). The Examiner concludes that one of skill would recognize the ability to substitute compounds that have the same GRA activity and have a reasonable expectation of success.

Bradley and Gebhard teach particular GRA compounds. Such disclosure does not cure the defects in the Examiner's arguments based on Schatzberg.

Claim 15 stands rejected under 35 USC § 103(a) as allegedly unpatentable over Schatzberg, in view of Belanoff.

The rejection of claim 15 in the September 30, 2008 Office Action included a summary of the rejection based on Schatzberg (as described under "Rejection" above). On page 8, the Examiner states that Schatzberg does not teach a specific GRA, and cites Belanoff for this information. According to the Examiner, Belanoff teaches that mifepristone is a specific GRA. The Examiner concludes that it would be obvious to employ a specific GRA for amelioration of the symptoms of PPP, motivated by the teaching of Schatzberg that GRAs ameliorate psychosis.

Appellants again note that the Examiner fails to acknowledge the teaching of Schatzberg as a whole. Schatzberg teaches that GRAs are effective for amelioration of psychoses *associated with glucocorticoid regulatory dysfunction.*

Nothing in Schatzberg, or Belanoff, or any other source cited by the Examiner suggests that PPP is associated with glucocorticoid regulatory dysfunction. The Exhibits submitted during prosecution demonstrate that the causes of PPP were not understood at the time, and that the art recognized that psychiatric disorders with similar symptoms cannot always be treated the same. Accordingly, it would not be obvious to one of skill at the time of the invention that a GRA, or a specific GRA, would be effective for treating PPP.

B. GROUND OF REJECTION 2 (Claims 3 and 4)

Claims 3 and 4 stand rejected under the first paragraph of 35 USC § 112 as allegedly lacking written description.

Claim 3 reads as follows:

> The method of claim 1, wherein the glucocorticoid receptor antagonist comprises a steroidal skeleton with at least one phenyl-containing moiety in the 11-β position of the steroidal skeleton.

Claim 4 reads as follows:

> The method of claim 3, wherein the phenyl-containing moiety in the 11-β position of the steroidal skeleton is a dimethylaminophenyl moiety.

This rejection presents the question of what constitutes sufficient disclosure for a class of chemical compounds, when the compound is not at the point of novelty of the invention. Here, the issue is whether art-recognized words and a functional definition can sufficiently describe such compounds. The GRA compounds disclosed in the specification, and in claims 3 and 4, are described using an internationally-recognized, standardized naming system, namely, IUPAC nomenclature. Exemplary compounds that fall within the scope of claims 3 and 4 are also described in the specification using IUPAC nomenclature.

Rejection

The Written Description rejection arose in the first Office Action from the present Examiner on September 30, 2009. On page 3, the Examiner set forth the issues as (1) what is meant by a "steroidal skeleton" and (2) what is meant by a "phenyl-containing moiety" and a "dimethyl aminophenyl moiety." The rejection continues:

> Further, the IUPAC definition of a moiety is 'half of a molecule including substructures of functional groups'. It is unclear to the Examiner if there is another part of the moiety that is undisclosed or if the other half of the moiety is the 'steroidal skeleton'.

According to the Examiner, the specification does not provide adequate support by such descriptive means as words, structures, figures, diagrams and formula that fully set forth the GRAs described in claims 3 and 4.

Appellants noted that words can be used to provide descriptive support for chemical compounds; there is no requirement for structural drawings. Appellants also submitted

evidence from the IUPAC guidelines demonstrating that the terms in claims 3 and 4 are recognized as descriptive of particular chemical structures. This was done in part because the Examiner cited IUPAC in the initial rejection, indicating an acknowledgement of the organization as an art-recognized authority.

Page 15 of the Final Office Action issued May 19, 2009, included the following comments:

> Page 11 of the instant specification states that 'the two most commonly known classes of structural modifications of the cortisol steroid backbone to create GRAs include modifications to the 11-β hydroxyl group'. One is left to theorize and conjecture as to which modifications are included or excluded by the recitation of 'a steroidal skeleton, 'at least one phenyl-containing moiety' and what is included or excluded in said moiety.

According to the Examiner, it would require undue, unpredictable experimentation to practice the claimed invention (May 19, 2009 Office Action, page 14).

Legal standard for written description

As set forth in MPEP 2163, written description requires that the specification describe the claimed invention in sufficient detail that one skilled in the art can reasonably conclude that the inventor had possession of the claimed invention. Thus, as with obviousness, the standard for written description is based on the point of view of one of skill in the art.

An applicant can show possession by describing the claimed invention with all of its limitations using such descriptive means as **words**, structures, figures, diagrams, and formulas that fully set forth the claimed invention. A common theme in the biological arts is whether there is a known correlation between the structure of a given compound and its function. A compound can be described functionally and comply with the written description requirement as long as there is some art-recognized correlation with the associated function. *See* MPEP 2163.

Further, the case law establishes that in certain circumstances, a claim can validly recite a class of compounds defined primarily by a shared function. These circumstances generally arise (i) when the point of novelty of the claim does not reside in the compounds

themselves, (ii) the class of compounds is known, and readily detectable by a routine assay, and (iii) there are a number of compounds that can be successfully used.

An example is found in *Ex parte Barrick*, 84 USPQ 142 (BPAI 1949). In *Barrick*, the claims at issue were directed to a process for preparing monomeric fluorine compounds, and the inventive step involved the use of a "polymerization inhibitor" in combination with a structurally defined composition. The Board held that the use of a "polymerization inhibitor" was acceptable because polymerization inhibitors were known, and because their use in the reaction was the point of novelty -- not the compounds themselves.

In re Fuetterer, 138 USPQ 217 (CCPA 1963) provides another example. The claims were directed to a rubber stock for making tires. The composition included several elements, including an inorganic salt with colloid suspending properties. The relevant language was as follows:

> ...an inorganic salt that is capable of holding a mixture of said carbohydrate and protein in colloidal suspension ... in an amount sufficient to hold the mixture ... in a film of water...

The specification included a few examples of salts that could be used. The court dismissed the argument that determining the suitability of undisclosed salts would constitute undue experimentation. The court noted that the invention was not found in the discovery that certain inorganic salts have colloid suspending properties, but that the exact point of novelty was the new combination of substances constituting the rubber tire tread stock.

The CCPA also considered the term "steroidal agent," recited in a claim to a method of enhancing membrane penetration using DMSO (*In re Herschler*, 200 USPQ 711 (CCPA 1979)). The specification included only one example of a steroidal agent used in the claimed method. The court found that the invention lay in the discovery that DMSO was useful for potentiating membrane penetration, not in the known chemical compound recited in "a manner auxiliary to the invention." According to the opinion, the written description need only be "so specific as to lead one of skill to that class of compounds."

The Examiner bears the burden of establishing a *prima facie* case by explaining why a person skilled in the art at the time the application was filed would not have recognized

that the inventor was in possession of the invention as claimed in view of the disclosure of the application as filed. MPEP 2163 II.A.2 states that the Examiner must review the entire specification prior to determining whether the disclosure satisfies the written description requirement.

The point of novelty is treatment of PPP with a GRA, not the recited steroidal GRAs

The present specification, as well as the claims, make clear that the invention is directed to treatment of a particular condition by inhibiting glucocorticoid signaling. The invention is based in part on the discovery that PPP is associated with dysfunctional glucocorticoid regulation, not on the discovery of particular GRAs. The identity of the GRA compound used to inhibit glucocorticoid signaling is less important than the fact that a universe of such compounds exists.

The GRAs recited in claims 3 and 4 represent a known class of compounds, as evidenced the references cited in the specification that describe these GRAs (*see* pages 10-12). The activity of a given compound as a GRA can be tested as described in the section entitled "Identifying Specific Glucocorticoid Receptor Antagonists," beginning on page 14.

GRAs having a phenyl-containing moiety at the 11-β position of the steroidal backbone are not novel, and do not represent the point of novelty of the present claims. The description provided, using art-accepted terminology, is sufficiently specific to lead one of skill to that class of compounds.

The specification describes GRA compounds, and provides a structure function correlation

The specification does not stop by defining glucocorticoid receptor antagonists functionally. A sizable portion of the disclosure is dedicated to describing various types of GRA compounds.

Steroidal GRA compounds are described in the section starting on page 10, line 17, which points to various modifications that can be made to the cortisol steroid structure to generate a GRA. Page 10, lines 19-21, states:

> Steroidal antiglucocorticoids can be obtained by modification of the basic structure of glucocorticoid agonists, i.e., varied forms of the steroid backbone. **The structure of cortisol can be modified in a variety of ways.** *(emphasis added)*

On page 11, lines 22-25, the specification states:

> For example, when bound to a 11-beta phenyl-aminodimethyl steroid, **the steroid receptor is maintained** in a conformation that cannot bind its natural ligand, such as cortisol in the case of glucocorticoid receptor.

These disclosures indicate that modifications can be made to the steroidal structure that preserve the essential structure of the natural ligand for the glucocorticoid receptor, *i.e.*, cortisol. There is no mystery as to what the "other half" of the molecule may be.

Possession can be demonstrated by words

GRA compounds, including the steroidal GRAs recited in claims 3 and 4, are described using the nomenclature standardized by IUPAC, an authority cited by the Examiner in the September 30, 2008 Office Action. This nomenclature was recognized in the art, at the time of filing, as descriptive of chemical structures. It is unclear to Appellants why a chemist or pharmacologist would not understand what is meant by a "steroidal skeleton" or a "phenyl-containing moiety," as recited in the claims.

As explained above, the specification describes the recited compounds in terms of function (glucocorticoid receptor antagonists), correlated with structure (similar to cortisol, but with a modification at a defined position on the steroidal skeleton). The specification also discloses three specific examples of steroidal compounds with phenyl groups at the 11-β position of the steroidal skeleton. The compounds are described on page 11-12 according to their common names, *i.e.*, RU486, RU009, and RU040, as well as according to IUPAC nomenclature, *i.e.*, 17-beta-hydrox-11-beta-(4-dimethyl-aminophenyl)17-alpha-(1-propynyl)estra-4,9-dien-3-one); 11-beta-(4-dimethyl-aminoethoxyphenyl)-17-alpha-(propynyl-17 beta-hydroxy-4,9-

estradien-3-one); and 17-beta-hydrox-17-alpha-19-(4-methyl-phenyl)-androsta-4,9 (11)-dien-3-one), respectively.

If the skilled artisan is left with any doubt that the inventors understood the nature of their invention, and the GRAs recited in claims 3 and 4 in particular, the specification points to several references that describe these modified steroid compounds (*see* page 10, line 17, to page 12, line 10).

The Examiner initially paraphrased the definition of moiety as "half of a molecule," and stated that it was unclear if there is another part of the moiety (half of a molecule) that is undisclosed or if the other half of the moiety (half of a molecule) is the steroidal skeleton. From this, it was unclear if the Examiner was referring to additional modifications to the steroidal skeleton, or to the phenyl-containing moiety.

For the purposes of clarification, Appellants submitted the IUPAC definition for moiety in the Response of January 28, 2009. **Ex. H**, a printout from the IUPAC Compendium of Chemical Technology, defines a moiety as "a part of a molecule." This was acknowledged by the Examiner in the May 19, 2009 Office Action.

In the same response, Appellants submitted evidence that the term "steroidal skeleton" would be "immediately envisioned" by one of skill at the time of the invention. **Ex. I** is an excerpt from "Definitive Rules for Nomenclature of Steroids," published by IUPAC in 1971. Pages 287-288 describe numbering of the carbons in the backbone, and α and β orientation. The exhibit demonstrates that the nomenclature for steroids was established as early as 1971, indicating that the term "steroidal skeleton," and positions on the steroidal skeleton do not require further description.

The Examiner has not applied the proper standard for written description

The Examiner has not considered the disclosure as a whole in rejecting claims 3 and 4. The Examiner pointed to a general disclosure on page 11 that discloses a class of GRAs, those with structural modifications to the 11-β hydroxyl group of the cortisol steroid backbone. The Examiner indicated it was unclear what else was included.

As explained above, the specification goes on from that point to include more detail about the structure of steroidal GRAs with modifications at the 11-β position. These compounds are designed to bind to the GR in a manner similar to cortisol in order to block interaction with the natural ligand. In addition, examples of steroidal GRAs that fall within the scope of claims 3 and 4 are provided.

The Examiner also seems to apply the standard for enablement by stating that it would require undue, unpredictable experimentation to practice the claimed invention. The CCPA cautioned against this approach in *Fuetterer* – there is no requirement that all undisclosed species be tested in the claimed method. Moreover, this is not the standard for written description. Regarding enablement, Appellants point again to the section entitled "Identifying Specific Glucocorticoid Receptor Antagonists," beginning on page 14, as enabling. This section describes a number of straightforward assays that can be used to rapidly screen steroidal compounds with an 11-β phenyl moiety for GRA activity.

The Examiner has yet to explain why one of skill would not immediately envision a steroidal compound with an 11-β phenyl moiety, given the familiar IUPAC nomenclature. The description of the recited steroidal compounds is sufficiently specific to lead one of skill to the recited class of compounds.

8. CONCLUSION

For these reasons, it is respectfully submitted that the rejections under 35 USC § 103 and under the first paragraph of 35 USC § 112 for written description should be reversed.

Respectfully submitted,

Carol P. Johns
Reg. No. 50,463

TOWNSEND and TOWNSEND and CREW LLP
Two Embarcadero Center, Eighth Floor
San Francisco, California 94111-3834
Tel: 415-576-0200
Fax: 415-576-0300

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9. CLAIMS APPENDIX

1. A method of ameliorating the psychotic symptoms of a patient having postpartum psychosis, comprising administering an amount of a glucocorticoid receptor antagonist effective to ameliorate the psychotic symptoms of the postpartum psychosis, with the proviso that the first psychotic symptoms arise within nine months of childbirth, that the patient has never suffered any psychotic condition not triggered by childbirth, and that the patient did not suffer from psychosis prior to parturition.

2. The method of claim 1, wherein the first psychotic symptoms arise within eight weeks of childbirth.

3. The method of claim 1, wherein the glucocorticoid receptor antagonist comprises a steroidal skeleton with at least one phenyl-containing moiety in the 11-β position of the steroidal skeleton.

4. The method of claim 3, wherein the phenyl-containing moiety in the 11-β position of the steroidal skeleton is a dimethylaminophenyl moiety.

5. The method of claim 4, wherein the glucocorticoid receptor antagonist comprises mifepristone.

6. The method of claim 4, wherein the glucocorticoid receptor antagonist is selected from the group consisting of 11β-(4-dimethylaminoethoxyphenyl)-17α-propynyl-17β-hydroxy-4,9 estradien-3-one and 17β-hydroxy-17α-19-(4-methylphenyl)androsta-4,9(11)-dien-3-one.

7. The method of claim 1 wherein the glucocorticoid receptor antagonist is selected from the group consisting 4α(S)-Benzyl-2(R)-prop-1-ynyl-1,2,3,4,4α,9,10,10α(R)-octahydro-phenanthrene-2,7-diol and 4α(S)-Benzyl-2(R)-chloroethynyl-1,2,3,4,4α,9,10,10α(R)-octahydro-phenanthrene-2,7-diol.

8. The method of claim 1, wherein the glucocorticoid receptor antagonist is (11β,17β)-11-(1,3-benzodioxol-5-yl)-17-hydroxy-17-(1-propynyl)estra-4,9-dien-3-one.

9. The method of claim 1, wherein the administration is once per day.

10. The method of claim 1, wherein the mode of administration is oral.

11. The method of claim 1, wherein the mode of administration is by a transdermal application, by a nebulized suspension, or by an aerosol spray.

15. The method of claim 1, wherein the glucocorticoid receptor antagonist is a specific glucocorticoid receptor antagonist.

10. EVIDENCE APPENDIX

A. Declaration under 37 C.F.R. § 1.132 by Joseph Belanoff, M.D.
Filed with Appellants' October 24, 2007 Response to a non-final Office Action.

B. Excerpts from Diagnostic and Statistical Manual of Mental Disorders, Fourth Ed. (DSM-IV), *published by* American Psychiatric Assoc. (2000).
Filed with Appellants' February 13, 2008 Response to a non-final Office Action.

C. Excerpts from Kaplan & Sadock's Comprehensive Textbook of Psychiatry, Seventh Ed., *published by* Lippencott Williams & Wilkins (2000).
Filed with Appellants' February 13, 2008 Response to a non-final Office Action.

D. Printout of Peace and Healing Postpartum Psychosis website, *available at* http://www.peaceandhealing.com/psychosis/postpartum.asp (last visited October 9, 2009).
Filed with Appellants' February 13, 2008 Response to a non-final Office Action.

E. Printout of NPR article "One Mother's Story," *available at* http://www.npr.org/programs/morning/features/2002/feb/postpartum/020218.postpartum.html (last visited October 9, 2009).
Filed with Appellants' February 13, 2008 Response to a non-final Office Action.

F. Elendov *et al.* (2001) *J. Clin. Endocrinol. Metab.* 86:4933-38
Filed with Appellants' January 28, 2009 Response to a Final Office Action.

G. Hendrick *et al.* (1998) *Psychosomatics* 39:93-101
Filed with Appellants' January 28, 2009 Response to a Final Office Action.

H. Printout from IUPAC Compendium of Chemical Terminology, definition of moiety, *available at* http://old.iupac.org/publications/compendium/index.html (last visited October 15, 2009).

Filed with Appellants' January 28, 2009 Response to a Final Office Action.

I. Excerpt from "Definitive Rules for Nomenclature of Steroids," published by IUPAC (1971), pages 287-288.

Filed with Appellants' January 28, 2009 Response to a Final Office Action.

11. RELATED PROCEEDINGS APPENDIX

This appeal has no related proceedings.

I hereby certify that this correspondence is being deposited with the United States Postal Service as first class mail in an envelope addressed to:

Assistant Commissioner for Patents
Washington, D.C. 20231

On January 20, 2000

By: [signature]

PATENT
Atty. Docket No.(TTC): 19904-002-1US
Client Reference (Stanford) No. 97-104
Attorney Docket No.(BFF)

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of:	Examiner: William Jarvis
SCHATZBERG & BELANOFF	Art Unit: 1614
Application No.: 09/244,457	
Filed: February 4, 1999	DECLARATION OF DR. JOSEPH BELANOFF UNDER 37 C.F.R. §1.132
For: METHODS FOR TREATING PSYCHOSIS ASSOCIATED WITH GLUCOCORTICOID RELATED DYSFUNCTION	

Assistant Commissioner for Patents
Washington, D.C. 20231

Sir:

I, Dr. Joseph Belanoff, being duly warned that willful false statements and the like are punishable by fine or imprisonment or both, under 18 U.S.C. § 1001, and may jeopardize the validity of the patent application or any patent issuing thereon, state and declare as follows:

1. All statements herein made of my own knowledge are true and statements made on information or belief are believed to be true. Exhibit 1, attached hereto, is incorporated herein by reference.

2. I received an M.D. in 1992 from Columbia University, College of Physicians and Surgeons.

3. I am presently employed at Stanford School of Medicine where I am conducting research to improve medical treatment for people suffering from psychosis. I am also the CEO of Corcept, Inc., whose primary mission is to provide improved medicine for psychiatric illnesses.

4. I have read and am familiar with the contents of the application. I understand that the Examiner has a rejection under §103 based upon his belief that one of skill reading the prior art of Ravaris, van der Lely, Piazza *et al.*, and Behl *et al.*, would have a reasonable expectation that patients with psychotic major depression [PMD] would be treatable with a glucocorticoid type II receptor antagonist.

5. We have conducted clinical trials which demonstrate that the invention was unpredictable. This unpredictability is demonstrated in two clinical trials that we conducted and that were reported to the Food and Drug Administration by way of an Annual Progress Report filed pursuant to an Investigational New Drug Application. A copy of that report is attached as **Exhibit 1**. In this report, the glucocorticoid receptor antagonist, mifepristone, is cited as being effective for treating psychosis in patients suffering from psychotic major depression and as having no clinical benefit for psychotic patients suffering from schizoaffective disease. The clinical studies were conducted by myself or at my direction.

6. Mifepristone for treating Psychotic Major Depression [PMD].

This study established that mifepristone was effective in treating psychosis in 4 out of 5 patients with the least psychotic patient being the least amenable to treatment. While all patients had improvement in the HAM-D depression test, the patients remained depressed and the improvement was attributed to the reduction in the level of psychosis.

The subjects were five newly admitted patients with an admitting diagnosis of major depression with psychotic features (DSM-IV criteria). The diagnosis at admission was confirmed independently by two psychiatrists. The subjects served as their own controls in a cross-over design. They were given either 600mg of mifepristone for four days, followed by four days of placebo; *or*, four days of placebo, followed by 600mg of mifepristone. Both the patients and the investigators were blind to which compound the patient was receiving. Routine biological and hematological studies were conducted daily in order to watch for evidence of relative adrenal insufficiency, such as hypoglycemia and eosinophilia.

The subjects had to be between the ages of eighteen and seventy-five, and without major medical problems. Patients were excluded if they had any signs of the Cushing Syndrome. Furthermore, because mifepristone in the dose range we used is reported to cause an abortion rate approaching eighty-five percent, women of childbearing potential were excluded from the study. All patients who admitted to having used illicit drugs within the month prior to admission, or who consumed in excess of two ounces of alcohol daily were also excluded.

Patients did not take anti-psychotic medication within three days of entering the study. No patients were taking antidepressant medication at the time they entered the mifepristone trial. No patient was *started* on an antidepressant medication while participating in the study. Benzodiazepines were permitted for insomnia and acetaminophen for headaches. If a patient's condition was such that they could not tolerate the drug-free period (for example, if they were intensely suicidal), they were

not studied. Finally, all patients were required to give written consent to a protocol approved by the Institutional Review Board at Stanford University Medical Center.

Formal psychiatric assessments (including the Hamilton Rating Scale for Depression[1] (HAM-D), Brief Psychiatric Rating Scale (BPRS) which measures psychosis, and Clinical Global Impression (CGI)), were carried out on days one, three, five, seven, and nine at 1000. On days one, five, and nine, paragraph recall was tested at 1130. Cortisol levels were measured serially every half-hour from 1300 to 1600, and plasma ACTH and plasma HVA were measured serially every hour from 1300 to 1600. Blood samples were spun down and plasma were frozen at -80°F in the General Clinical Research Center Laboratory. Plasma cortisol determinations were made by radioimmunoassay (RIA) in the Endocrinology Laboratory at Brigham and Women's Hospital (Harvard University). Plasma ACTH was assayed by immunoradiometric assay (IRMA) in the same laboratory.

FIGURE 1: EXAMPLE TIMELINE FOR A PATIENT WHO RECEIVED MIFEPRISTONE FIRST (DOUBLE BLIND STUDY).

DAY 1	
10:00	Psychiatric Assessments (HAM-D, BPRS, CGI)
11:30	Paragraph Recall
13:00-16:00	Afternoon Cortisol Test
13:00-16:00	Plasma ACTH & HVA

[1] Hamilton M: A Rating Scale for Depression. *J Neurol Neurosurg Psychiat* 1960; 23:56-62.

Day 2	Subject started on 600mg mifepristone
Day 3 10:00	600mg mifepristone Psychiatric Assessments (HAM-D, BPRS, CGI)
Day 4	600mg mifepristone
Day 5 10:00 11:30 13:00-16:00 13:00-16:00	600mg mifepristone Psychiatric Assessments (HAM-D, BPRS, CGI) Paragraph Recall Afternoon Cortisol Test Plasma ACTH & HVA
Day 6	600mg Placebo
Day 7 10:00	600mg Placebo Psychiatric Assessments (HAM-D, BPRS, CGI)
Day 8	600mg Placebo
Day 9 10:00 11:30 13:00-16:00 13:00-16:00	600mg Placebo Psychiatric Assessments (HAM-D, BPRS, CGI) Paragraph Recall Afternoon Cortisol Test Plasma ACTH & HVA

BRIEF PATIENT HISTORIES

PATIENT 1

This 50-year-old man had no prior psychiatric history, and had received no mental health treatment except for "career counseling" in graduate school. He was employed as an executive in the high-tech industry, was in excellent physical health, and was married with no children. He took no medications other than daily vitamins. Three months prior to his entry into the study he noted increasing feelings of depression with anhedonia, insomnia, decreased appetite, and decreased concentration. A stressor at that time was his mother's entry into a skilled nursing facility because of advanced

Alzheimer's Disease. One month prior to entry into the study, he began to grow increasingly suspicious that co-workers were talking about him and "planning to get him fired." At entry into the study, he was extremely guarded with mood-congruent delusions that the hospital might be a prison where he would be executed. He had received no psychiatric care to that point.

At admission, the subject's mean afternoon cortisol level was 12.0μg/dL, and did not decline throughout the afternoon collection period. He received mifepristone first, and by day 5 his mean afternoon cortisol level was 37.7μg/dL; and, in a striking example, the normal rhythm of a steady decline of cortisol levels throughout the afternoon had resumed (see Table 1). His HAM-D declined from 29 to 21, and his BPRS declined from 47 to 40. Moreover, from day 5 to day 9, while on placebo, his HAM-D continued to drop (21 to 10), as did his BPRS (40 to 25), suggesting that mifepristone continued to be active in his system, as indicated by the continued elevation of his afternoon cortisol values. At this time, his normal cortisol rhythm continued. The patient experienced no adverse effects and no lab values other than cortisol, and ACTH changed significantly. The patient was started on the antidepressant, paroxetine, at discharge and returned to work two weeks later. His depressed mood resolved over the next several weeks and his paroxetine was discontinued nine months after conclusion of the study. He remains asymptomatic two years later.

Table 1: Results of the Afternoon Cortisol Test (Patient 1)

Time	Day 1 Cortisol Levels (μg/dL)	Day 5 Cortisol Levels (μg/dL)	Day 9 Cortisol Levels (μg/dL)
1300	11.8	56.0	22.1
1330	14.4	40.9	22.7
1400	11.6	34.4	16.9
1430	10.4	34.2	14.1
1500	11.6	34.6	13.4
1530	12.7	35.7	12.6
1600	11.8	28.4	18.7
Mean	12.0μg/dL	37.7μg/dL	17.2μg/dL

PATIENT 2

This subject was a 44 year-old European-American woman with a history of one prior episode of PMD; three years prior to study admission, she had been hospitalized for one week with florid symptoms of depression and psychosis. During her initial episode of PMD she acknowledged being very depressed and felt that the devil was controlling her. She knew this to be true because her bed was very cold and there "might have been a machine under [her] bed." Against medical advice, she left the hospital because she came to believe that one of her physicians was also being controlled by the devil. After leaving the hospital, she continued to be severely depressed with both auditory hallucinations as well as somatic delusions. She tried paroxetine for several weeks but there was no change in her condition. The paroxetine was decreased and nortriptyline was started. Eventually, lithium was added to her treatment regimen, and her depression improved, although her somatic delusions remained.

One year prior to study admission she found that all of her symptoms had resolved. Two months later, against medical advice, she decided to discontinue taking medications. For nine months she remained asymptomatic, but then became depressed again. She could not identify any particular precipitating event. She reported increasingly depressed mood, weight loss, decreased concentration, memory, and energy, anhedonia, and insomnia.

One month after the onset of this depressive episode she attempted suicide by hanging. The attempt failed because her feet reached the floor. She then made a second suicide attempt by taking an overdose of the previously prescribed nortriptyline. At this point, she was brought to the Emergency Department and stabilized. During her exam, she revealed that she had recently been hearing strange noises in her house and "seeing shadows." She also stated that "the devil [was] manipulating her body" and that she had been unwilling to drive because "the devil [had] the power to destroy her." Her exam in the Emergency Department was also notable for significant psychomotor retardation.

Prior to her first hospitalization, the subject had no psychotic history. She immigrated to the U.S. in 1992, has been married for 21 years, has two children, and works as a domestic. Her only long-standing medical problems are irritable bowel syndrome and back pain. Her family history includes one sister who suffers from "mood swings," and her father who suffers from alcoholism. At the time of study entry, the subject's physical exam was within normal limits, and she was on estradiol and medroxyprogesterone for perimenopausal symptoms.

The subject received placebo first, and then mifepristone. While on placebo, her HAM-D increased from 33 to 35 and her BPRS from 51 to 57. While on mifepristone, her HAM-D declined from 35 to 21 and her BPRS from 57 to 44. At the end of the nine-day study, the subject was no longer delusional and felt well enough to go home. She declined follow-up antidepressant medication. Six weeks after leaving the study, she was reported to be suffering from symptoms of PMD and did not return for follow-up.

PATIENT 3

The subject, a 67 year-old woman with a history of recurrent PMD, was admitted after taking 15 fluoxetine capsules in a suicide attempt. Her first episode of PMD was in 1980 during which she suffered from delusions of persecution and reference, and was hospitalized following a suicide attempt. One year prior to study entry she suffered from an episode of PMD and was prescribed a low dose of haloperidol and fluoxetine. Her condition improved to the point where she felt "back to normal," and after 2 to 3 months of combination therapy she decided to stop taking her prescribed haloperidol and fluoxetine. Two months prior to study admission, her condition began to deteriorate. She complained of very low energy, poor appetite, spontaneous crying, poor self-care, and increased guilt about being a burden to her family. She also expressed increasingly delusional thoughts including that her phones were tapped, her family was trying to poison her, her neighbors were observing her through her windows; and, most recently, that white automobiles were following her. There was a question of whether she suffered from auditory hallucinations because she

complained of hearing "sirens" and "phones ringing," but this observation was complicated by her partial hearing loss.

The subject's psychiatric history has been marked by long periods when she is fully functional (working as a nursing aide) with intermittent episodes of severe depression and paranoid ideation. At admission, the subject was taking no medications of any kind on a regular basis. Other than a 65% hearing loss in one ear, and a 35% loss in the other ear, she had no ongoing medical problems.

This subject received placebo first and then mifepristone. While on placebo her HAM-D declined from 23 to 19 and her BPRS increased from 32 to 35. While on mifepristone, her HAM-D declined from 19 to 17 and her BPRS increased from 35 to 36. She was discharged on olanzapine and her condition continued to improve. Her mean afternoon cortisol was 9.4μL/dL at entry into the study, 9.4μL/dL after four days of placebo, greater than 60μL/dL after four days of mifepristone, but only 3.1μL/dL when she returned for a follow-up eight weeks later and was feeling well.

PATIENT 4

This subject was a 57 year-old male physician with an 18-month history of severe depression characterized by extreme insomnia, low energy, poor concentration, and somatic concerns that had resulted in an extensive medical work-up. Despite an extraordinary physical workup, he could not be convinced that he was physically sound and planned even more extensive physical testing. He tried virtually all of the antidepressants currently available, often in combination with antipsychotic

medication. He also had a round of ECT therapy, with 8 treatments that led to a mild diminution of symptoms that quickly faded. He had not been able to work for the past 15 months, which was in sharp contrast to a very productive career prior to the onset of his depression. He linked the onset of his depression to treatment with prednisone for an allergic reaction. He had no prior history of depression and no medical problems, but his family history was significant for his mother having severe late-life depression. He had weaned himself of all medications (with the exception of clonazepam for sleep) prior to study entry.

The subject received placebo first. While on placebo his HAM-D declined from 31 to 28 and his BPRS declined from 53 to 45. While on mifepristone his HAM-D declined from 28 to 21 and his BPRS from 45 to 28. He left the hospital at the end of the study and was started on venlafaxine, a treatment to which his depression had not previously responded. Although his course of recovery was not a straight line, his improvement continued over time and he required neither further hospitalization nor ECT to eventually gain full recovery.

PATIENT 5

This subject was a 45 year-old man with a history of obsessive-compulsive disorder. In the 8 months prior to study entry, he became increasingly depressed, suffering from poor sleep, anhedonia, poor concentration, low energy, and feelings of guilt, and had developed a fixed belief that his hearing had been irreparably harmed by various noises in his environment. These noises included a phone ringing, a child's bell, and a car horn. He became convinced that he had lost "almost all" of his

hearing and was not dissuaded by the many trips to the audiologist, which indicated normal hearing, nor by the fact that he could converse in normal tones with those around him. Several weeks before study admission, he contemplated suicide and was briefly involuntarily hospitalized because he was a danger to himself. After trying a first dose of several medications, he refused to take any medication because he believed that each previous one or two pill trials had added to his hearing loss. Shortly before study admission, he began to "sense" that the police were "trailing him" ever since his involuntary admission. This subject worked as a college professor, and was married with three children. He used no illicit substances or alcohol, but did have a family history of several siblings with major depression, his mother suffered from both depression and dementia prior to her death.

This subject received mifepristone first and then placebo. While on mifepristone, his HAM-D declined from 46 to 37 and his BPRS from 54 to 41. Of note is that item 11 (suspiciousness) declined from a "6", severe, to a "1", absent, and item 15 (unusual thought content) declined from a "6" to a "3", mild. He no longer believed that the "police [were] trailing him" nor that his phone was tapped. However, he still obsessively believed that he had a hearing loss, and his desire to have his hearing re-tested was even stronger than before. While on placebo his HAM-D declined from 37 to 35, but his BPRS increased again from 41 to 54, with particularly high scores on "somatic concern" and anxiety. At discharge he refused all medications, and he has remained quite debilitated with high levels of somatic anxiety.

Results

Table 2: Individual HAM-D Scores

Subject #	Day 1	Day 5	Day 9
1 (mifepristone first)	29	21	10
2 (placebo first)	33	35	21
3 (placebo first)	23	19	17
4 (placebo first)	31	28	21
5 (mifepristone first)	46	37	35

In all cases, HAM-D scores declined during mifepristone treatment. In both cases, where the subject received mifepristone first, their HAM-D declined farther during the placebo treatment than if placebo received first (case one significantly, case five marginally). In the three cases where placebo was given first, HAM-D scores changed very little (rising slightly in case two and falling slightly in cases three and four.) Ignoring the carryover effect leaves five active treatment cells and three placebo cells. The mean decline in HAM-D while on mifepristone was 8.4 (31%) while on placebo it was 1.2 (7%). The difference approaches statistical significance ($F = 5.01$, $p<.07$).

Table 3: Individual BPRS Scores

Subject #	Day 1	Day 5	Day 9
1 (mifepristone first)	49	40	25
2 (placebo first)	51	57	44
3 (placebo first)	32	35	36
4 (placebo first)	53	45	28
5 (mifepristone first)	54	41	54

In all cases but one, BPRS scores declined during mifepristone treatment. (The exception was case three, the patient with the lowest BPRS at study

entry. Her BPRS score increased by one point.) In one case where the subject received mifepristone first (case one) their BPRS continued a distinct decline during the placebo period. In the other subject who received mifepristone first (case 5), the subject's BPRS reversed to the pretreatment level during placebo treatment. The mean decline in BPRS score was 10.6 points (32.5%) while on mifepristone, while BPRS increased .3 points (.5%) while the subjects were on placebo. The difference again approaches statistical significance ($F = 4.31$, $p<.08$).

Conclusion: All the patients were discharged from the hospital at the end of the nine-day study period. All of the five subjects showed significant improvement in their HAM-D scores while on mifepristone and four of the five subjects share improvement in their BPRS scores. Moreover, the subject who did not was the least symptomatic to start. The 32.5% overall decline in BPRS approaches the 40% value frequently seen in six to eight week trails of effective antipsychotic medication. None of the subjects reported side effects of any kind, and both basic lab measures and measures of vital signs were unaffected by treatment.

While HAM-D [psychosis] scores diminished during treatment, all the patients still had significant residual signs and symptoms of major depression. We recommend that all patients begin antidepressant treatment at the end of the study. We observed that the more significant clinical change was that 4 of the 5 patients were no longer psychotic at the end of the study and all were more cognitively organized. The reason that the patients' HAM-D scores declined was apparently due to their being more cognitively intact and because they felt in better control of their thinking.

7. Schizoaffective disorder tests did not work.

As reported in the attached letter (Exhibit 1) to the FDA, we did a double-blind, placebo controlled clinical trial for patients having been diagnosed with schizoaffective disorder. This disease describes a patient who is suffering from both schizophrenia and mood disorders, most typically depression. In this study, both psychotic patients suffered from depression. The results of the study are provided below in Table 4. All scoring was done by blinded raters who were not staff members.

In the first round of the trial, both patients were given placebo and no effect on their BPRS scores were noted. After completion of the study, the patients were given the option and elected to try mifepristone in an open-label study. Despite the increased likelihood that the patients would respond because they and the staff knew that they were receiving mifepristone, neither patient demonstrated any improvement in BPRS.

Table 4
TREATMENT OF SCHIZOAFFECTIVE DISORDER USING MIFEPRISTONE

Subject No.	Scale	Day 1 (raw score)	Day 9 (raw score)	Δ
1	HAM-D	24	17	-7
(placebo)	BPRS	47	53	+6
	CGI	4 (moderately depressed)	5 (markedly depressed)	+1
2	HAM-D	33	30	-3
(placebo)	BPRS	51	46	-5
	CGI	4 (moderately depressed)	4	0

OPEN-LABEL STUDY OF THE TREATMENT OF SCHIZOAFFECTIVE DISORDER USING MIFEPRISTONE

Subject No.	Scale	Day 1 (raw score)	Day 9 (raw score)	Δ
1	HAM-D	15	15	0
	BPRS	40	39	-1
	CGI	3 (mildly depressed)	4 (moderately depressed)	+1
2	HAM-D	28	21	-7
	BPRS	50	49	-1
	CGI	4 (moderately depressed)	4	0

This Declarant has nothing further to say.

Dated: January 11, 2000

Joseph Belanoff, M.D.

Attachment: Exhibit I

TOWNSEND and TOWNSEND and CREW LLP
Two Embarcadero Center, 8th Floor
San Francisco, California 94111-3834
Tel: (415) 576-0200
Fax: (415) 576-0300
KAW/jhd

SF 1041100 v1

Stanford University School of Medicine

Department of Psychiatry & Behavioral Sciences
Stanford, California 94305-5717

(650) 723-6811
FAX (650) 498-5294

IND 50,269

July 16, 1999

Paul Leber, M.D.
Division Director
Food and Drug Administration
Division of Neuropharmacological Drug Products (HFD-120)
Woodmont 2
1451 Rockville Pike
Rockville, MD 20852

SUMMARY: Annual Report of Progress – IND# 50,269 (mifepristone)

Investigational Study Titles: "Rapid reversal of psychotic depression using mifepristone"
"Treatment of schizoaffective disorder using mifepristone"

Number of Patients Treated to Date: 8

Most Recent Subject Enrollment Date: July 12, 1999

Adverse Reactions Encountered: none.

Modifications to the Protocol(s): none.

Dear Dr. Leber:

We continue to use mifepristone in our protocols "Rapid reversal of psychotic major depression using mifepristone (RU 486)" and "Treatment of schizoaffective disorder using mifepristone." To date, we have been impressed by its relative lack of side effects and its potential efficacy. We have received no subjective complaints from any of the research subjects while they were participating in our protocols nor have we found any adverse effects in frequently administered tests of physiological function while subjects were on protocol.

You will find a summary of our clinical data and results on page two of this letter. Our group has not presented this data in any formal presentations or papers. This data continues to be confidential. If you have any questions regarding this report, please contact Joseph K. Belanoff, M.D., at (650) 725-5586.

Sincerely,

Alan F. Schatzberg, M.D.
Kenneth T. Norris, Jr., Professor and Chairman
Department of Psychiatry and Behavioral Sciences
Stanford University School of Medicine
401 Quarry Road
Stanford, CA 94305-5717

AFS/brs

EXHIBIT 1

Clinical Data and Results

STUDY TITLE: "Rapid reversal of psychotic depression using mifepristone"

Number of subjects successfully completing protocol = 5

This laboratory has observed in a current study that psychosis, altered cognition, and mood have all rapidly improved in patients with psychotic major depression when the glucocorticoid-receptor antagonist mifepristone was used for short periods. Our experimental protocol (in progress) calls for a four-day double-blind, placebo-controlled crossover study that uses 600 mg of mifepristone as the active agent. We have noted that because there is no intervening "washout period," there is a significant biochemical carryover effect.

In each of our first five cases, 21-item Hamilton Rating Scale for Depression[1] (HAM-D) scores declined during the 4 days of mifepristone treatment, on average 25.5% (i.e., from an average score of 31.4 → 23.4 on the last day). HAM-D scores remained essentially unchanged when the subjects received placebo during the first arm (before crossover) of the study (29.0 → 27.3). In both cases where subjects received mifepristone first, their HAM-D scores continued to decline in the placebo arm, in one case slightly and in the other case significantly.

On average, Brief Psychiatric Rating Scale[2] (BPRS) scores declined by 10.2 points (a change of 34.0%) while subjects were taking mifepristone. In contrast, average BPRS scores for the subjects administered placebo first increased by 3 points (5%). In one case where the subject received mifepristone first, the BPRS score continued a significant decline during the placebo period; in the other case, the subject's BPRS score rose to the pre-treatment level by the end of the placebo period.

Average Clinical Global Impression (CGI) scores declined by 33.0% (a decrease of 1.2 points on a seven-point scale, i.e., "much improved" relative to baseline) while subjects were taking mifepristone. For those subjects receiving placebo first, CGI scores declined by 8.0% (or 0.3 points). (Please refer to following summary on page 3.)

STUDY TITLE: "Treatment of schizoaffective disorder using mifepristone"

Number of subjects successfully completing protocol = 2

This double-blind, placebo-controlled clinical trial calls for research subjects with a diagnosis of schizoaffective disorder to be assigned randomly to receive *either* 400 mg mifepristone or placebo for eight days. Both subjects studied to date were randomized to the placebo arm of this study. In addition, both subjects chose to receive active medication (on an open-label basis for eight days) when the blind was broken at the conclusion of the randomized study protocol.

HAM-D, BPRS, and CGI scores did not change significantly in either patient when they were taking placebo or open-label mifepristone. Please see following summary of patient data.

---

[1] Hamilton M: A rating scale for depression. *J Neurol Neurosurg Psychiatry* 1960; 23:56-62.
[2] Overall J, Gorham D: Brief Psychiatric Rating Scale. *Psychol Rep* 1962; 10:799.

Raw Data

RAPID REVERSAL OF PSYCHOTIC DEPRESSION USING MIFEPRISTONE (RU 486)

Subject No.	Scale	Day 1 (raw score)	Day 5 (raw score)	Day 9 (raw score)	Δ
1	HAM-D	29	21	10	-19
(RU486 first,	BPRS	49	40	25	-24
then placebo)	CGI	6 (severely depressed)	6	3 (mildly depressed)	-3
2	HAM-D	33	35	21	-12
(placebo first,	BPRS	51	57	44	-7
then RU486)	CGI	6 (severely depressed)	6	5 (markedly depressed)	-1
3	HAM-D	23	19	17	-6
(placebo first,	BPRS	32	35	36	+4
then RU486)	CGI	4 (moderately depressed)	4	3 (mildly depressed)	+1
4	HAM-D	31	28	21	-10
(placebo first,	BPRS	53	45	28	-25
then RU486)	CGI	7 (extremely depressed)	6 (severely depressed)	5 (markedly depressed)	-2
5	HAM-D	46	37	35	-11
(RU486 first,	BPRS	54	41	54	0
then placebo)	CGI	5 (markedly depressed)	6 (severely depressed)	4 (moderately depressed)	-1
6	HAM-D	41	35	--	-6
(unknown,	BPRS	49	42	--	-7
in progress)	CGI	6 (severely depressed)	6	--	0

TREATMENT OF SCHIZOAFFECTIVE DISORDER USING MIFEPRISTONE

Subject No.	Scale	Day 1 (raw score)	Day 9 (raw score)	Δ
1	HAM-D	24	17	-7
(placebo)	BPRS	47	53	+6
	CGI	4 (moderately depressed)	5 (markedly depressed)	+1
2	HAM-D	33	30	-3
(placebo)	BPRS	51	46	-5
	CGI	4 (moderately depressed)	4	0

OPEN-LABEL STUDY OF THE TREATMENT OF SCHIZOAFFECTIVE DISORDER USING MIFEPRISTONE

Subject No.	Scale	Day 1 (raw score)	Day 9 (raw score)	Δ
1	HAM-D	15	15	0
	BPRS	40	39	-1
	CGI	3 (mildly depressed)	4 (moderately depressed)	+1
2	HAM-D	28	21	-7
	BPRS	50	49	-1
	CGI	4 (moderately depressed)	4	0

DIAGNOSTIC AND STATISTICAL MANUAL OF MENTAL DISORDERS

FOURTH EDITION

TEXT REVISION

DSM-IV-TR™

Published by the
American Psychiatric Association
Washington, DC

Mood Disorders

The Mood Disorders section includes disorders that have a disturbance in mood as the predominant feature. The section is divided into three parts. The first part describes mood episodes (Major Depressive Episode, Manic Episode, Mixed Episode, and Hypomanic Episode) that have been included separately at the beginning of this section for convenience in diagnosing the various Mood Disorders. These episodes do not have their own diagnostic codes and cannot be diagnosed as separate entities; however, they serve as the building blocks for the disorder diagnoses. The second part describes the Mood Disorders (e.g., Major Depressive Disorder, Dysthymic Disorder, Bipolar I Disorder). The criteria sets for most of the Mood Disorders require the presence or absence of the mood episodes described in the first part of the section. The third part includes the specifiers that describe either the most recent mood episode or the course of recurrent episodes.

The Mood Disorders are divided into the Depressive Disorders ("unipolar depression"), the Bipolar Disorders, and two disorders based on etiology—Mood Disorder Due to a General Medical Condition and Substance-Induced Mood Disorder. The Depressive Disorders (i.e., Major Depressive Disorder, Dysthymic Disorder, and Depressive Disorder Not Otherwise Specified) are distinguished from the Bipolar Disorders by the fact that there is no history of ever having had a Manic, Mixed, or Hypomanic Episode. The Bipolar Disorders (i.e., Bipolar I Disorder, Bipolar II Disorder, Cyclothymic Disorder, and Bipolar Disorder Not Otherwise Specified) involve the presence (or history) of Manic Episodes, Mixed Episodes, or Hypomanic Episodes, usually accompanied by the presence (or history) of Major Depressive Episodes.

Major Depressive Disorder is characterized by one or more Major Depressive Episodes (i.e., at least 2 weeks of depressed mood or loss of interest accompanied by at least four additional symptoms of depression).

Dysthymic Disorder is characterized by at least 2 years of depressed mood for more days than not, accompanied by additional depressive symptoms that do not meet criteria for a Major Depressive Episode.

Depressive Disorder Not Otherwise Specified is included for coding disorders with depressive features that do not meet criteria for Major Depressive Disorder, Dysthymic Disorder, Adjustment Disorder With Depressed Mood, or Adjustment Disorder With Mixed Anxiety and Depressed Mood (or depressive symptoms about which there is inadequate or contradictory information).

Bipolar I Disorder is characterized by one or more Manic or Mixed Episodes, usually accompanied by Major Depressive Episodes.

Bipolar II Disorder is characterized by one or more Major Depressive Episodes accompanied by at least one Hypomanic Episode.

Criteria for Atypical Features Specifier

Specify if:

With Atypical Features (can be applied when these features predominate during the most recent 2 weeks of a current Major Depressive Episode in Major Depressive Disorder or in Bipolar I or Bipolar II Disorder when a current Major Depressive Episode is the most recent type of mood episode, or when these features predominate during the most recent 2 years of Dysthymic Disorder; if the Major Depressive Episode is not current, it applies if the feature predominates during any 2-week period)

A. Mood reactivity (i.e., mood brightens in response to actual or potential positive events)

B. Two (or more) of the following features:

(1) significant weight gain or increase in appetite
(2) hypersomnia
(3) leaden paralysis (i.e., heavy, leaden feelings in arms or legs)
(4) long-standing pattern of interpersonal rejection sensitivity (not limited to episodes of mood disturbance) that results in significant social or occupational impairment

C. Criteria are not met for With Melancholic Features or With Catatonic Features during the same episode.

Postpartum Onset Specifier

The specifier With Postpartum Onset can be applied to the current (or, if the full criteria are not currently met for a Major Depressive, Manic, or Mixed Episode, to the most recent) Major Depressive, Manic, or Mixed Episode of Major Depressive Disorder, Bipolar I Disorder, or Bipolar II Disorder or to Brief Psychotic Disorder (p. 329) if onset is within 4 weeks after childbirth. The symptoms of the postpartum-onset Major Depressive, Manic, or Mixed Episode do not differ from the symptoms in nonpostpartum mood episodes. Symptoms that are common in postpartum-onset episodes, though not specific to postpartum onset, include fluctuations in mood, mood lability, and preoccupation with infant well-being, the intensity of which may range from overconcern to frank delusions. The presence of severe ruminations or delusional thoughts about the infant is associated with a significantly increased risk of harm to the infant.

Postpartum-onset mood episodes can present either with or without psychotic features. Infanticide is most often associated with postpartum psychotic episodes that are characterized by command hallucinations to kill the infant or delusions that the infant is possessed, but it can also occur in severe postpartum mood episodes without such specific delusions or hallucinations. Postpartum mood (Major Depressive, Manic, or Mixed) episodes with psychotic features appear to occur in from 1 in 500 to 1 in 1,000 deliveries and may be more common in primiparous women. The risk of post-

partum episodes with psychotic features is particularly increased for women with prior postpartum mood episodes but is also elevated for those with a prior history of a Mood Disorder (especially Bipolar I Disorder). Once a woman has had a postpartum episode with psychotic features, the risk of recurrence with each subsequent delivery is between 30% and 50%. There is also some evidence of increased risk of postpartum psychotic mood episodes among women without a history of Mood Disorders with a family history of Bipolar Disorders. Postpartum episodes must be differentiated from delirium occurring in the postpartum period, which is distinguished by a decreased level of awareness or attention.

Women with postpartum Major Depressive Episodes often have severe anxiety and even Panic Attacks. Maternal attitudes toward the infant are highly variable but can include disinterest, fearfulness of being alone with the infant, or overintrusiveness that inhibits adequate infant rest. It is important to distinguish postpartum mood episodes from the "baby blues," which affect up to 70% of women during the 10 days postpartum, are transient, and do not impair functioning. Prospective studies have demonstrated that mood and anxiety symptoms during pregnancy, as well as the "baby blues," increase the risk for a postpartum Major Depressive Episode. A past personal history of nonpostpartum Mood Disorder and a family history of Mood Disorders also increase the risk for the development of a postpartum Mood Disorder. The risk factors, recurrence rates, and symptoms of postpartum-onset Mood Episodes are similar to those of nonpostpartum Mood Episodes. However, the postpartum period is unique with respect to the degree of neuroendocrine alterations and psychosocial adjustments, the potential impact of breast-feeding on treatment planning, and the long-term implications of a history of postpartum Mood Disorder on subsequent family planning.

Criteria for Postpartum Onset Specifier

Specify if:

With Postpartum Onset (can be applied to the current or most recent Major Depressive, Manic, or Mixed Episode in Major Depressive Disorder, Bipolar I Disorder, or Bipolar II Disorder; or to Brief Psychotic Disorder)

Onset of episode within 4 weeks postpartum

Specifiers Describing Course of Recurrent Episodes

A number of specifiers for Mood Disorders are provided to increase diagnostic specificity and create more homogeneous subgroups, assist in treatment selection, and improve the prediction of prognosis. Specifiers that describe the course of recurrent episodes include Longitudinal Course Specifiers (With and Without Full Interepisode Recovery), Seasonal Pattern, and Rapid Cycling. These specifiers cannot be coded. Table 2 indicates which course specifiers apply to each Mood Disorder (see p. 424).

Kaplan & Sadock's

COMPREHENSIVE TEXTBOOK of PSYCHIATRY

SEVENTH EDITION

Editors

BENJAMIN J. SADOCK, M.D.

VIRGINIA A. SADOCK, M.D.

VOLUME ONE

LIPPINCOTT WILLIAMS & WILKINS

▲ 13.4 Postpartum Psychiatric Syndromes

RUTA NONACS, M.D., PH.D. and LEE S. COHEN, M.D.

ond postpartum week require further evaluation to rule out the evolution of a more serious affective illness.

Postpartum Depression Major depressive disorder is relatively common during the postpartum period. Both retrospective and prospective community-based studies have revealed rates of postpartum minor and major depression in the range of 10 to 15 percent. These rates of depression reported in puerperal cohorts are similar to those observed in nonpuerperal populations of women.

While some women report the acute onset of symptoms shortly after delivery, depression more commonly develops insidiously over the first 6 postpartum months. A significant proportion of women actually experience the onset of depressive symptoms during pregnancy. The signs and symptoms of postpartum depression are generally indistinguishable from those characteristic of nonpsychotic major depressive disorder that occurs in women at other times. Dysphoric mood, irritability, anhedonia, insomnia, and fatigue are frequently reported; somatic complaints are also common. Ambivalent or negative feelings toward the infant are often reported, and it is common for a woman with postpartum depression to express doubts or concerns about her ability to care for her child. In its most severe form, postpartum depression may result in profound dysfunction. Suicidal ideation is frequently reported; however, suicide rates appear to be relatively low in women who become depressed during the postpartum period.

Although few studies have evaluated the prevalence of comorbid psychiatric illness in this population, severe anxiety and obsessionality are prominent in women with puerperal illness. Symptoms of generalized anxiety, panic disorder, and obsessive-compulsive disorder are often observed in women with postpartum depression.

Puerperal Psychosis Puerperal psychosis is the most severe form of postpartum psychiatric illness. In contrast to postpartum blues and depression, puerperal psychosis is a rare event that occurs in approximately 1 to 2 per 1000 women after childbirth. Its presentation is often dramatic, with onset of psychosis as early as the first 48 to 72 hours postpartum. Most women with puerperal psychosis develop symptoms within the first 2 to 4 weeks after delivery.

In women with this disorder, psychotic symptoms and disorganized behavior are prominent and cause significant dysfunction. Puerperal psychosis resembles a rapidly evolving affective psychosis with manic, depressive, or mixed features. The earliest signs are typically restlessness, irritability, and insomnia. Women with this disorder typically exhibit a rapidly shifting depressed or elated mood, disorientation or depersonalization, and disorganized behavior. Delusional beliefs often center on the infant and include delusions that the child may be defective or dying, that the infant has special powers, or that the child is either Satan or God. Auditory hallucinations that instruct the mother to harm or kill herself or her infant are sometimes reported. Although most believe that this illness is indistinguishable from an affective (or manic) psychosis, some have argued that puerperal psychosis may be clinically distinct in that it is more commonly associated with confusion and delirium than nonpuerperal psychotic mood disorder.

Postpartum Blues Many women experience mild depressive symptoms during the first week after delivery, which are commonly known as postpartum blues or "baby blues." Depending on the criteria used to diagnose the blues, prevalence estimates range from 30 to 85 percent. Women with postpartum blues report a variety of symptoms, including dysphoria, mood lability, irritability, tearfulness, anxiety, and insomnia. These symptoms typically peak on the fourth or fifth day after delivery and remit spontaneously by the tenth postpartum day. Postpartum blues are relatively benign and are, by definition, time-limited. While the occurrence of postpartum blues does not necessarily reflect psychopathology in the mother, some women with blues will go on to develop postpartum depression. Women with histories of mood disorder require close monitoring, as some data suggest that blues may herald the development of major depressive disorder in women who have had previous episodes of affective illness. Symptoms of the blues that persist beyond the sec-

Peace and Healing

with Dan Williams Psy. D., PA-C

Postpartum Psychosis

Post-partum psychosis is very rare. It is not so much a variety of post-partum depression as it is an entity onto itself. It is characterized by homicidal and suicidal impulses, hallucinations, delusions, disorganized and bizarre thinking.

The dilemma is that these individuals usually refuse treatment. This is a medical emergency situation. If post-partum psychosis is suspected, families need to call 911 as emergency intervention is necessary. Medication most likely will be prescribed. The ultimate goal is to keep the baby and mother safe.

Research shows that approximately one woman in 1,000 births will experience post-partum psychosis.

As with treating any psychosis, we need to first rule out any physiological cause such as thyroid storms, seizure disorders or drug-induced psychosis. Once ruled out, treatment is crucial. However before individual or group therapy, medication must be given to alleviate the psychotic symptomatolgy. In many cases, anti-psychotic medication is administered only for a short period of time. Once the psychosis abates, couple counseling, as well as individual counseling, can be of benefit

Family support, and educating the family regarding what has occurred is also extremely important. It is important that the affected individual not be labeled a bad mother. Once the psychosis is treated, mothers generally go on to be good caretakers. It is important to note, however, that in the event of future pregnancies, affected individuals are at a 50% greater risk of having another psychotic episode.

Certain anti-psychotics have been more affective than others. Haldol, Risperidal, Clozaril and Zyprexa have been beneficial in post-partum psychosis.

Anti-psychotic medications do pass into the mother's breast milk. Subsequently if the mother has been breastfeeding and continues to do so, the baby needs to be monitored for drowsiness or lethargic behavior, and prescribing the least amount of anti-psychotic medication in order for symptom reduction to occur is also crucial.

EXHIBIT
D
1 Page Total

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One Mother's Story

Postpartum Psychosis: Rare, Frightening and Treatable

Listen to Joanne Silberner's story.

After narrowly conquering her own bout of postpartum psychosis, Shelley Ash became a health professional so she could help others with the disorder. *Photo: Dan Ash*

Feb. 18, 2002 -- Today in Houston, a jury will begin hearing testimony in the murder trial of Andrea Yates. She's accused of drowning her five young children last June. Her lawyers have said she'll plead not guilty by reason of insanity, citing her history of postpartum depression with psychosis. For *Morning Edition*, NPR's Joanne Silberner reports that the condition is rare, frightening -- and treatable.

When Shelley Ash of San Jose, Calif., was pregnant five years ago, she read all the baby books she could. She never came across the term postpartum psychosis. Then she gave birth to her son.

"I knew right away something was wrong," says Ash. She sensed she was watching the delivery from above. She was terrified. Hospital nurses told her the feeling would pass. It didn't, even after she and her baby went home.

Ash says was pacing all the time, and caught in a horrible depression. She was constantly crying, couldn't sleep and couldn't eat.

Symptoms of Postpartum Psychosis

Postpartum psychosis is a more rare and severe disorder than postpartum depression. It affects about 1 in 500 to 1,000 new mothers. Onset is severe and quick, and should be treated as a medical emergency. Symptoms include:

Delusions, or false beliefs.

This wasn't the "baby blues," a temporary anxiety and depression that hits about three-quarters of mothers. Nor was it the postpartum depression that afflicts one in 10 new mothers. As Ash eventually learned, she was suffering from postpartum psychosis, which hits about one in 500 to one in a 1,000 mothers within three months of birth.

"Postpartum psychosis is condition in which the person

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loses touch with reality," says Dr. Ralph Wittenberg, who runs a postpartum screening project in Washington, D.C. Mothers hear voices, see things and feel an irrational guilt that they've somehow done something wrong, he says. Without treatment, women may try to hurt themselves or those around them.

What was happening to Ash was beyond her control. Postpartum psychosis sometimes develops out of postpartum depression, or hits women who've had previous psychiatric problems. Sometimes, as in Ash's case, it shows up out of the blue. Psychiatrists aren't sure what causes such a sudden and powerful break with reality, but they believe the changing hormones and stress of childbirth are somehow involved.

Ash knew she was getting worse. The midwife in her obstetrician's office told her to call a psychiatrist. But that was the last thing Ash wanted to do.

"I was terrified," says Ash. She was having delusions and was afraid that if she told anyone about what she was thinking or seeing in her mind, they would take her son away.

But after one episode, she became desperate.

She remembers watching David Letterman drop watermelons from high places on his television show. "But that turned into my son," she says. "I kept imagining how it would be to drop him out of his bedroom window and he would go splat on the pavement below and shatter into a million pieces."

The image was too much for her. Ash went to her bathroom cabinet and took an overdose of painkillers she had been

Symptoms of Postpartum Depression

About 70 percent of new mothers get the "baby blues" -- feelings of anxiety and irritability that can hit three or four days after delivery, but disappear quickly. Postpartum depression, which can appear even a year after giving birth, is more severe and can last for months, if not treated. About 1 in 10 new mothers experience the disorder. Symptoms include:

Severe sadness or emptiness; emotional numbness or apathy.
Withdrawal from family, friends or pleasurable activities.
Constant fatigue, trouble sleeping, overeating or loss of appetite.
A strong sense of failure or inadequacy.
Intense worry about the baby or a lack of interest in the baby.
Hallucinations -- hearing voices or seeing things that are not real.
Thoughts of harming or killing the baby.
Unwillingness to eat or sleep.
Frantic energy.
Severe depressive symptoms.

Thoughts about suicide; fears of harming the baby.

prescribed for a previous back injury.

Her husband came home from a run to find her on the floor in the front hall, babbling, and he rushed her to the hospital. That pattern isn't unusual, says Dr. Nada Stotland, who specializes in women's health. Stotland says women with postpartum psychosis tend to know that something's wrong, but like Ash, they're often terrified to let anyone else know. Health professionals and family members need to recognize that delusions and really erratic behavior are sure signs of trouble, and that hospitalization and medication are needed, says Stotland.

Ash was hospitalized for a few days. She then spent 18 months on anti-psychotic, anti-depressant and anti-anxiety medications. Her son is now 5 years old and healthy. Ash got a graduate degree in public health so she could educate people about postpartum psychosis. She couldn't be happier now about her own health, or the health of her son. But she's not having any more children.

It's not worth the risk of getting sick again, she says. There's a 10 to 20 percent chance that without treatment, she would, according to British research. But Stotland and others say that with treatment at the first sign of a problem, women with a history of postpartum psychosis can safely have more babies. And while neither psychiatrist would comment on Andrea Yates -- the woman who killed her five children -- both say that the tragedy whenever a case of postpartum psychosis ends badly is that most likely it could have been prevented.

Join the Discussion

Postpartum depression and psychosis often go unrecognized and untreated. Weigh in with your thoughts or experiences with the disorders at NPR's discussion board.

In Depth

» Browse for more NPR stories about postpartum depression.

Other Resources

• Postpartum Support International is a nonprofit resource and support center sponsored in part by the Indiana University of Pennsylvania. Visit its Web site for listings for support groups worldwide, online chat rooms and bulletins, book recommendations and information on postpartum illness.

• Read a factsheet on postpartum illness, provided by the Office on Women's Health.

• Visit Depression After Delivery, Inc., a national, nonprofit organization, for other resources, including telephone support and referral services for women and families coping with mental health

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IL-12, TNF-α, and Hormonal Changes during Late Pregnancy and Early Postpartum: Implications for Autoimmune Disease Activity during These Times

ILIA J. ELENKOV*, RONALD L. WILDER*, VLADIMIR K. BAKALOV, AMREY A. LINK, MARIANA A. DIMITROV, SCOTT FISHER, MARIANNA CRANE, KEITH S. KANIK, AND GEORGE P. CHROUSOS

Arthritis and Rheumatism Branch (I.J.E., R.L.W., M.C., K.S.K.), National Institute of Arthritis and Muscul Skin Diseases, Pediatric and Reproductive Endocrinology Branch (I.J.E., A.A.L., S.F., G.P.C.), National Ins Health and Human Development, Developmental Endocrinology Branch (V.K.B.), National Institute of Chil Human Development, and Laboratory of Experimental and Computational Biology (M.D.), National Cancer National Institutes of Health, Bethesda, Maryland 20892

Clinical observations indicate that some autoimmune diseases, such as rheumatoid arthritis and multiple sclerosis, frequently remit during pregnancy but exacerbate, or have their onset, in the postpartum period. The immune basis for these phenomena is poorly understood. Recently, excessive production of IL-12 and TNF-α was causally linked to rheumatoid arthritis and multiple sclerosis. We studied 18 women with normal pregnancies in their third trimester and during the early postpartum period. We report that during the third trimester pregnancy, *ex vivo* monocytic IL-12 production was about 3-fold and TNF-α production was approximately 40% lower than postpartum values. At the same time, urinary cortisol and norepinephrine excretion and serum levels of 1,25-dihydroxyvitamin were 2- to 3-fold higher than postpartum values. As shown previously, these hormones can directly suppress IL-12 and TNF-α production by monocytes/macrophages *in vitro*. We suggest that a cortisol-, norepinephrine-, and 1,25-dihydroxyvitamin-induced inhibition and subsequent rebound of IL-12 and TNF-α production may represent a major mechanism by which pregnancy and postpartum alter the course of or susceptibility to various autoimmune disorders. (*J Clin Endocrinol Metab* 86: 4933–4938, 2001)

IL-12, PRODUCED by antigen-presenting cells, is a major inducer of T helper 1 (Th1) responses by stimulating Th1 lymphocyte proliferation and differentiation and by inducing interferon (IFN)-γ production from natural killer and T cells (1, 2). Antigen-presenting cell-derived IL-12 and TNF-α, in concert with Th1 cell-derived IFN-γ, stimulate the activity of T cytotoxic and natural killer cells, and monocytes/macrophages, *i.e.* the major components of cellular immunity. IL-12 and TNF-α are considered major proinflammatory cytokines because they stimulate the synthesis of nitric oxide and other inflammatory mediators that drive chronic delayed-type inflammatory responses (2). On the other hand, the antiinflammatory cytokine IL-10 produced by monocytes/macrophages and Th2 cells promotes humoral immunity and inhibits monocyte/macrophage activation and the production of proinflammatory cytokines (1). Excessive production of IL-12 and TNF-α and a deficit of IL-10 appears to play a key role in the inflammatory activity and the tissue damage observed in organ-specific autoimmune diseases, such as rheumatoid arthritis (RA) and multiple sclerosis (MS) (3–5). Moreover, excessive IL-12 production is the pivotal factor in the proliferation and differentiation of pathogenic autoreactive Th1 effector cells in the experimental models of these diseases (6).

Some autoimmune diseases, such as RA and MS, often remit during pregnancy, particularly in the third trimester, but have an exacerbation or their onset during the postpartum period (7–10). The risk of developing new onset RA during pregnancy, compared with nonpregnancy, is decreased by about 70%. In contrast, the risk of developing RA is markedly increased in the postpartum period, particularly the first 3 months (odds ratio of 5.6 overall and 10.8 after first pregnancy) (10). Moreover, a substantial fraction (20–30%) of premenopausal onset RA develops within 1 yr of pregnancy (R. Wilder, unpublished observations). In women with multiple sclerosis, the rate of relapse declines during pregnancy, especially in the third trimester, increases during the first 3 months postpartum, and then returns to the prepregnancy rate (8). Although documented extensively, these observations remain poorly understood.

The third trimester of pregnancy and the early postpartum period are also known to be associated with abrupt changes of several hormones, including in tandem increases and decreases, respectively, of E2, progesterone, cortisol, and 1,25-dihydroxyvitamin D_3 (9, 11). Recently, we and others demonstrated that cortisol, catecholamines (norepinephrine and epinephrine), and 1,25-dihydroxyvitamin D_3 are potent inhibitors of IL-12 and TNF-α production by monocytes/macrophages *ex vivo* and *in vitro* (12–16). We hypothesized that during late pregnancy the increase of these hormones, and their rapid decline in the early postpartum period, may induce opposite changes in both IL-12 and TNF-α production (17). Therefore, we examined the production of IL-12 and TNF-α after lipopolysaccharide (LPS) stimulation of whole

Abbreviations: IFN, Interferon; LPS, lipopolysaccharide; MS, multiple sclerosis; NE, norepinephrine; RA, rheumatoid arthritis; Th, T helper.

Exhibit F 6 pages total

blood cultures *ex vivo* and measured the levels of E2, progesterone, cortisol, 1,25-dihydroxyvitamin D_3, and catecholamines in women during gestation wk 33–36 and 3–6 wk after delivery.

Materials and Methods

Subjects

Eighteen healthy pregnant women between the ages of 20 and 40 yr and 18 age-matched, healthy, nonpregnant women participated in the study, which was approved by the institutional review board of the NIH. The pregnant women underwent testing during gestation wk 33–36 and 3–6 wk after delivery. They were enrolled in the study with the approval of their obstetricians. We screened each participant at the NIH Clinical Center by history, physical examination, and routine laboratory tests. All signed informed consents. The controls had their tests during the early and mid follicular phases of their menstrual cycle (d 3–8). All participants abstained from taking medications (except prenatal vitamins and iron supplements in pregnancy) during the week before the study. Blood specimens for hormone measurements were drawn after 1 h of rest between 1300 and 1400 h. Urine samples were collected for two 24-h periods during the preceding 2 d to measure free cortisol and catecholamine excretion rates.

Whole blood cultures

Ex vivo whole blood cytokine production assays were performed as described elsewhere (12). Blood was drawn into sodium-heparin-containing sterile tubes (Vacutainer, Becton Dickinson and Co., Lincoln Park, NJ) and processed within 45 min. The blood, diluted 1:5 with RPMI 1640 (supplemented with 1% glutamine and 50 μg/ml gentamicin) with no added exogenous serum, was divided into aliquots (1.0 ml) in 24-well cell culture plates (Costar, Cambridge, MA). To induce cytokine production, bacterial LPS was added at 1 μg/ml final concentration, and the samples were incubated in 5% CO_2 at 37 C for 18 h. After incubation, the blood was centrifuged, and the supernatant plasma was separated and stored in polypropylene tubes at −70 C until assayed.

The whole blood *ex vivo* cytokine assay, which has recently found favor elsewhere (18), has several advantages. This method avoids the isolation of leukocytes from whole blood that may cause activation and artifactual differences not present *in vivo*. The method also preserves the "natural environment" (including hormones) of cytokine-producing cells. Importantly, in comparison to methods using isolated peripheral blood mononuclear cells, the whole blood assay also shows less intra-individual variation. Less than 15% intraindividual variation of whole blood cytokine production is reported when subjects are sampled over time (18, 19) (Elenkov, I. J., R. L. Wilder, and G. P. Chrousos, unpublished observations). This contrasts with the wide (but stable) range of IL-12, TNF-α, and IL-10 secretion levels seen across healthy individuals (interindividual variation) (18–21), demonstrating that this test forms a good basis for the study of genetically or hormonally defined variation.

Monocytes/macrophages are the main IL-12-, TNF-α-, and IL-10-producing cells in LPS-stimulated whole blood (22). In view of the observed changes of monocyte/macrophage numbers during pregnancy (see *Results*), the whole blood cytokine production was corrected for monocyte/macrophage counts (pg per 10^6 monocytes/macrophages).

Cytokine assays

IL-12 p70, TNF-α, and IL-10 were measured using ELISA employing the multiple antibody sandwich principle (Quantikine, R&D Systems, Inc., Minneapolis, MN). IL-12 p70 ELISA recognizes specifically the biologically active IL-12 heterodimer without cross-reactivity with the individual subunits of the dimer (p35 and p40). The detection limits of the IL-12 p70 and the high sensitivity IL-12 p70 ELISA were 7.5 and 0.5 pg/ml, respectively, whereas they were 15.0 and 2.0 pg/ml for the TNF-α and IL-10 ELISA. The quality control parameters of these ELISAs were as follows: intraassay coefficient of variation (CV), 1.1–1.5%; interassay CV, 3.3–7.1%. Plates were read by a microplate reader (model 550, Bio-Rad Laboratories, Inc., Richmond, CA), and absorbance was transformed to cytokine concentration (pg/ml) using a standard curve computed by Microplate Manager III (Macintosh Data Analysis Software, Bio-Rad Laboratories, Inc.).

Hormonal measurements

E2 was measured by RIA after extraction and LH20 column chromatography. Intraassay CV was 4.5%, and interassay CV was 11%. Normal values for the follicular phase are 0.38–.367 nmol/l. Progesterone was measured by RIA after extraction with hexane. Interassay CV was 6.7%, and intraassay CV was 4.5%. Normal values for the follicular phase are 0.003–0.03 nmol/l. 1,25-Dihydroxyvitamin D_3 was measured by cartridge extraction and RIA. Intraassay and interassay CVs were 10%. Normal values are 53–161 pmol/l (Mayo Clinical Laboratories, Rochester, MN). Twenty-four-hour urinary excretion of free cortisol was measured after extraction by chemiluminescent competitive protein binding assay. Intraassay and interassay CVs were 4.4%. Normal values are 66–298 nmol/24 h (Mayo Clinic Laboratories). Twenty-four-hour urinary excretion of epinephrine and norepinephrine (NE) were measured by HPLC with electrochemical detection. Intraassay and interassay CVs were 3.5 and 4.0, respectively. Normal values are 0–109 and 89–473 nmol/24 h, respectively (Mayo Clinic Laboratories).

Data analysis

All data are presented as means ± SE. ANOVA was done with Statistica (version 5.5, StatSoft, Inc., Tulsa, OK). Pregnancy and postpartum values of the same individuals were compared by repeated measures ANOVA. Values from healthy age-matched control subjects were compared with those of pregnancy and postpartum subjects using one-way ANOVA.

Results

Blood count changes during pregnancy and postpartum

Pregnancy was associated with an increase of white blood cell counts compared with healthy, nonpregnant controls and the postpartum state ($8.8 \pm 0.5 \times 10^3$ mm^3 *vs.* $6.0 \pm 0.3 \times 10^3$ mm^3, both in controls and postpartum; $P < 0.001$). This was attributable to a significant increase of polymorphonuclear leukocytes and monocytes (mean, $6.6 \pm 0.5 \times 10^3/\mu$l and $0.62 \pm 0.04 \times 10^3/\mu$l, respectively) during pregnancy compared with healthy matched nonpregnant controls (mean, $3.3 \pm 0.3 \times 10^3/\mu$l and $0.38 \pm 0.03 \times 10^3/\mu$l, respectively; $P < 0.001$) and the postpartum state (mean, $3.4 \pm 0.2 \times 10^3/\mu$l and $0.4 \pm 0.02 \times 10^3/\mu$l, respectively; $P < 0.001$). Pregnancy was also associated with a moderate but significant decrease ($P < 0.05$) of lymphocytes and eosinophil counts (mean, $1.8 \pm 0.1 \times 10^3/\mu$l and $0.09 \pm 0.01 \times 10^3/\mu$l, respectively) compared with control age-matched nonpregnant women (mean, $2.2 \pm 0.1 \times 10^3/\mu$l and $0.16 \pm 0.03 \times 10^3/\mu$l, respectively).

Decrease of IL-12 and TNF-α production during pregnancy

During pregnancy, whole blood IL-12 production was decreased 2-fold compared with the postpartum period (61.0 ± 10.5 *vs.* 120.7 ± 31.8 pg/ml, respectively). When corrected for monocyte count, the decrease of IL-12 production was more pronounced (>3 fold; Table 1). The individual changes of IL-12 production corrected for monocyte count in 18 women during pregnancy and their follow-up in the postpartum period are shown in Fig. 1. During pregnancy, 15 of the women had lower IL-12 production than postpartum. Of interest, we found a large interindividual variation of the "effect of pregnancy" on IL-12 production, *i.e.* 5 individuals

TABLE 1. Summary of LPS-induced IL-12 and TNF-α production *ex vivo* and hormone levels in 18 subjects during the third trimester of pregnancy and 3–6 wk after delivery and in 18 healthy age-matched nonpregnant women

Cytokine or Hormone	Nonpregnant age-matched controls	Pregnancy	Postpartum	Controls *vs.* pregnancy	Pregnancy *vs.* postpartum	Controls *vs.* postpartum
IL-12	142.1 ± 60.4	106.8 ± 21.4	340.1 ± 88.5	NS	<0.01	NS
TNF-α	5803.2 ± 971.6	5648.8 ± 484.5	7829.9 ± 870.4	NS	0.07	NS
24-h urinary free cortisol	109.1 ± 12.2	380.1 ± 31.9	134.2 ± 16.9	<0.001	<0.001	NS
24-h NE	184.9 ± 10.7	285.2 ± 17.4	132.6 ± 10.2	<0.001	<0.001	<0.01
25-dihydroxy-vitamin D	57.4 ± 6.3	87.4 ± 5.8	78.4 ± 4.6	<0.001	NS	<0.05
1,25-dihydroxy-vitamin D	111.9 ± 7.3	260.0 ± 20.6	94.1 ± 5.9	<0.0001	<0.0001	NS
E2	246.6 ± 29.2	53816.1 ± 11547.6	231.5 ± 151.9	<0.001	<0.001	NS
Progesterone	1.6 ± 0.4	380.1 ± 48.4	1.1 ± 0.4	<0.001	<0.001	NS

Data are presented as means ± SE. Cytokine production is corrected for monocyte number (pg/10^6 monocytes). Hormone levels are expressed as nmol/L, except for 1,25-dihydroxy-vitamin D, which is expressed in pmol/L; UFC (urinary free-cortisol) and urinary NE are expressed as nmol/24 h.

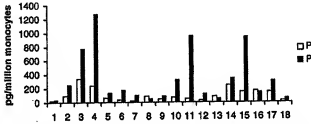

FIG. 1. Individual changes of IL-12 production by whole blood cultures stimulated with bacterial LPS *ex vivo* in 18 subjects during the third trimester of pregnancy and 3–6 wk after delivery. Cytokine production is corrected for monocyte number (pg/10^6 monocytes). P, Pregnancy; PP, postpartum.

had dramatic changes of IL-12 production, whereas others had moderate or minimal changes.

The production of TNF-α per monocyte was decreased by 40% during pregnancy compared with the postpartum period and narrowly failed to reach statistical significance ($P = 0.07$; Table 1). However, we observed similar levels of TNF-α production in pregnant *vs.* nonpregnant women (Table 1). There were no significant differences of LPS-induced IL-10 production by monocytes among the control, pregnant, and postpartum groups (data not shown).

Normal pregnancy is characterized by marked hormonal changes

The 24-h urinary cortisol excretion was increased about 4-fold during pregnancy compared with the nonpregnant state (Table 1), and in all cases it exceeded the upper limit of the reference values. After delivery, the cortisol excretion returned to normal levels. No changes in 24-h urinary excretion of epinephrine and dopamine were observed (data not shown), but we found that the 24-h urinary NE excretion was significantly increased during pregnancy and returned to baseline or lower levels in the postpartum period (Table 1).

As expected, the serum levels of E2 and progesterone were markedly increased during pregnancy. Postpartum, the ovarian hormone levels returned to normal follicular phase levels (Table 1). During pregnancy, plasma 25-hydroxyvitamin D_3 was increased by 50%, whereas 1,25-dihydroxyvitamin D_3 increased more than 2-fold (Table 1).

Cytokine production and hormone levels in a single individual before, during, and after pregnancy

We had the opportunity to follow the cytokine and hormonal changes of one woman (subject 4 in Fig. 1) as nonpregnant, during pregnancy, and postpartum (Fig. 2). She had a substantial decline in IL-12 production during pregnancy compared with the nonpregnant state. Three weeks after delivery, when cortisol, NE, E2, progesterone, and 1,25-dihydroxyvitamin D_3 returned to prepregnancy levels or lower, there was a notable rebound of LPS-induced IL-12 and TNF-α production.

E2 and progesterone do not affect IL-12, TNF-α, and IL-10 production

No data are available regarding whether E2 and progesterone are able to modulate the production of IL-12 by monocytes/macrophages. The significant changes of E2 and progesterone during pregnancy prompted us to study their direct effects in our assay system. Neither E2 nor progesterone at 10^{-5} to 10^{-11} M modulated the production of IL-12, TNF-α, or IL-10 in the LPS-stimulated human whole blood from five normal, nonpregnant individuals and three pregnant individuals (data not shown).

Discussion

We demonstrated that during late pregnancy, compared with the postpartum period, the capacity of monocytes to produce IL-12 was reduced more than 3-fold, whereas the capacity for TNF-α production was reduced by ~40%. The decreased production of these proinflammatory cytokines was paralleled by significant increases of cortisol, NE, 1,25-dihydroxyvitamin D_3, E2, and progesterone.

The pregnant women also had lower LPS-induced IL-12 production compared with age-matched controls, although the difference did not reach statistical significance (Table 1). The lack of significance in this case may reflect the large interindividual variability of monocytic IL-12 production across healthy individuals (Table 1). However, we observed a clear suppression of IL-12 production during pregnancy and a rebound in the postpartum when we followed a single individual through the nonpregnant, pregnant, and postpartum states (Fig. 2). Thus, because of the substantial interindividual variability of IL-12 production, larger and more

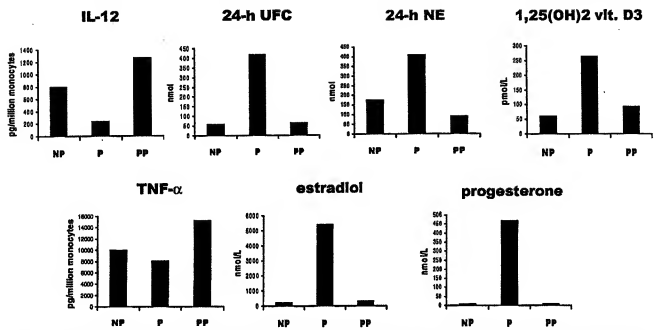

FIG. 2. Proinflammatory cytokine and hormonal changes in subject 4 (see Fig. 1) in the nonpregnant, pregnant, and postpartum states. Cytokine production is corrected for monocyte number (pg/10^6 monocytes), and hormone levels are expressed as nmol/l, except for 1,25-dihydroxyvitamin D_3, which is expressed as pmol/l. NP, Nonpregnant state; P, pregnancy; PP, postpartum; UFC, urinary free cortisol.

extended longitudinal studies are needed to address the differences between pregnancy and the nonpregnacy state.

Like others (23), we did not observe a difference of TNF-α production in pregnant *vs.* nonpregnant women. This might reflect the same factors described above for IL-12 production and an increase of soluble TNF receptors p55 and p75 in the third trimester of pregnancy, documented by Russell *et al.* (23). We did not find significant differences of LPS-induced IL-10 production by monocytes among the control, pregnant, and postpartum groups. Previous studies have shown increased production of IL-10 by peripheral blood mononuclear cells and placenta during pregnancy (24, 25). This discrepancy with our observations most likely reflects methodological differences. In the LPS-stimulated whole blood assay used by us, the primary source of IL-10 is the monocyte (22), whereas in the isolated peripheral blood mononuclear cell assay, which involves stimulation by mitogens, the primary source of IL-10 most likely is the lymphocyte.

The substantially increased urinary free cortisol excretion during the third trimester of pregnancy that returned to normal levels 3 wk after delivery indicates that late pregnancy is a state of adrenocortical activation, probably caused by the large amounts of CRH secreted by the placenta (11). We also found a significant increase of 24-h NE urinary excretion during pregnancy with a return to low normal levels in the postpartum period. This is consistent with observations in pregnant rats, in which a more than 2-fold increase of 24-h urinary excretion of NE has been described (26). Further studies are needed to elucidate whether these observations are linked to increased sympathetic nerve activity and/or reduced NE uptake during pregnancy, although, most likely, both take place (see also below).

The moderate increase of serum 25-hydroxyvitamin D_3 during pregnancy probably reflects the increased levels of serum vitamin D-binding proteins (27, 28). Like Seely *et al.* (28), we observed a more than 2-fold increase of the highly regulated serum 1,25-dihydroxyvitamin D_3 in the third trimester of pregnancy. These changes most likely result from increased conversion of 25-hydroxyvitamin D_3 to 1,25-dihydroxyvitamin D_3 in the human placenta, in addition to the increase of vitamin D-binding proteins (27, 28).

Recent evidence indicates that glucocorticoids, NE, and 1,25-dihydroxyvitamin D_3 potently inhibit the production of IL-12 and TNF-α by human monocytes/macrophages *in vitro* and *ex vivo* (12–16, 29). These hormones also inhibit the production of IL-2 and IFN-γ by Th1 cells (15, 29). In contrast, glucocorticoids do not affect the production of IL-10 by monocytes, but they potentiate IL-10 and IL-4 production by Th2 cells (12, 29, 30). Thus, the observed hormonal changes during pregnancy may explain the inhibition of monocytic IL-12 and TNF-α production. Furthermore, because IL-12 is extremely potent in enhancing IFN-γ and inhibiting IL-4 synthesis by T cells, the inhibition of IL-12 production may represent an important mechanism by which these hormones mediate a Th2 shift during pregnancy (Fig. 3).

We did not demonstrate a direct effect of E2 or progesterone on the production of IL-12, TNF-α, or IL-10 by human monocytes *ex vivo*. However, estrogens may affect cytokine production indirectly by enhancing the activity of the stress system, *i.e.* via increases in the secretion of cortisol and catecholamines (11). In addition, estrogens are potent inhibitors of the extraneuronal uptake of NE (uptake 2) (31), which may also explain the increased NE excretion in pregnancy demonstrated in our study. Therefore, estrogens may amplify the IL-12/TNF-α-inhibitory and Th2-facilitatory activities of cor-

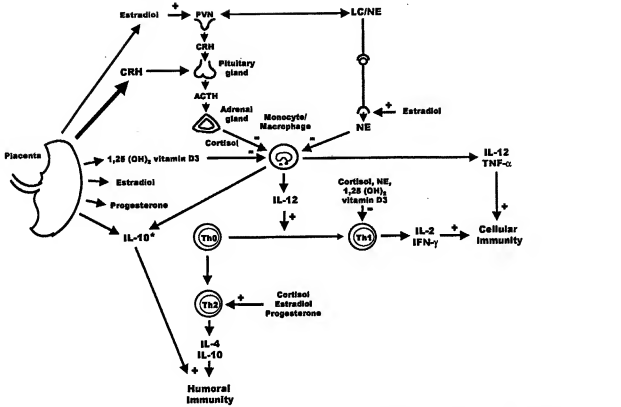

FIG. 3. A proposed simplified model of the role of different hormones in the regulation of innate and Th1 and Th2 cytokine profiles during pregnancy. Th1 cells primarily secrete IFN-γ and IL-2, which promote cellular immunity, whereas Th2 cells secrete primarily IL-4 and IL-10, which promote humoral immunity. During pregnancy, lymphocyte cytokine production is skewed toward the Th2 type: peripheral lymphocytes secrete less IFN-γ and IL-2 but more IL-4 and IL-10, particularly in the third trimester (24, 25). IL-12, a 75-kDa heterodimeric cytokine produced mostly by monocytes/macrophages, is a central inducer of Th1 responses and cell-mediated immunity by favoring Th1 cell proliferation and differentiation and by suppressing Th2 responses. Hypothalamic CRH stimulates the secretion of pituitary ACTH, which in turn triggers the secretion of cortisol from the adrenal cortex. During human pregnancy, the placenta is the major source of circulating CRH. The placenta also secretes IL-10 that may stimulate humoral and suppress cellular immunity. The sympathetic system innervates all peripheral tissues, including blood vessels and lymphoid organs. Upon activation, the sympathetic nerve terminals in these organs release NE locally and into the bloodstream. Cortisol, NE, 1,25-dihydroxyvitamin D_3, E2, and progesterone have multiple and divergent effects on the immune system. *Cortisol does not affect the production of IL-10 by monocytes/macrophages (see text). Note that *cortisol E2, and progesterone up-regulate IL-10 production by Th2 lymphocytes. In addition, E2 stimulates the activity of the CRH neurons and increases local NE concentrations by blocking its uptake. Thus, *in vivo*, E2 might amplify the effects of cortisol and NE. The net result of these complex hormonal effects is the suppression of IL-12 and TNF-α production, Th1 responses, and a Th2 shift. This hormonally induced Th2 shift may suppress Th1-related diseases such as RA and MS during pregnancy, whereas the rebound of IL-12 and TNF-α production and Th1 responses in the postpartum may facilitate the flares or the onset of these diseases. Note that several other factors, besides hormones (*e.g.* antibodies, soluble cytokine receptors, etc.), that most likely are also involved in the modulation of Th1/Th2 balance during pregnancy and postpartum, are not discussed here. LC, Locus ceruleus; PVN, paraventricular nucleus.

tisol and NE *in vivo* (Fig. 3). Furthermore, progesterone and estrogens up-regulate the production of IL-4 and IL-10 by Th2 cells *in vitro* (32, 33). Thus, an increase of estrogens and progesterone may also facilitate a Th2 shift during pregnancy by directly stimulating the production of IL-4 and IL-10 by Th2 cells (Fig. 3). This is consistent with recent data documenting increased IL-4 and IL-10 production by lymphocytes and the placenta during the third trimester of pregnancy (24, 25).

In conclusion, we demonstrated that human third trimester pregnancy, compared with the early postpartum period, is characterized by a reduction of the monocytic production of the Th1 type/proinflammatory cytokines IL-12 and TNF-α and by an increase of the secretion of cortisol, NE, and 1,25-dihydroxyvitamin D_3. Postpartum, when these hormones return to normal or low normal levels, the removal of their inhibitory effects may induce a rebound of IL-12 and TNF-α production and a Th1 shift.

The changes of Th1 type/proinflammatory cytokine production observed in this study may provide new understanding of the clinical observations that Th1-related diseases such as RA and MS frequently remit during pregnancy but exacerbate or have their onset in the postpartum period. Our study also suggests that some individuals have exaggerated postpartum Th1 type/proinflammatory cytokine rebound, raising the question of the factors that control this phenomenon. These individuals could be at greater than average risk for developing or exacerbating already existing

autoimmune diseases. Thus, further studies of the role of neuroendocrine factors in the regulation of IL-12, TNF-α/IL-10, and Th1/Th2 balance may suggest novel diagnostic and therapeutic approaches for these diseases.

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Address all correspondence and requests for reprints to: Dr. Ilia J. Elenkov, Division of Rheumatology, Immunology, and Allergy, Georgetown University Medical Center, 3800 Reservoir Road NW, Washington, D.C. 20007-2197. E-mail: ije@gunet.georgetown.edu.

* These authors contributed equally to this work.

References

1. Mosmann TR, Sad S 1996 The expanding universe of T-cell subsets: Th1, Th2 and more. Immunol Today 17:138–146
2. Trinchieri G 1995 Interleukin-12: a proinflammatory cytokine with immunoregulatory functions that bridge innate resistance and antigen-specific adaptive immunity. Annu Rev Immunol 13:251–276
3. Morita Y, Yamamura M, Nishida K, et al. 1998 Expression of interleukin-12 in synovial tissue from patients with rheumatoid arthritis. Arthritis Rheum 41:306–314
4. Matusevicius D, Kivisakk P, Navikas V, Soderstrom M, Fredrikson S, Link H 1998 Interleukin-12 and perforin mRNA expression is augmented in blood mononuclear cells in multiple sclerosis. Scand J Immunol 47:582–590
5. van Boxel-Dezaire AH, Hoff SC, van Oosten BW, et al. 1999 Decreased interleukin-10 and increased interleukin-12p40 mRNA are associated with disease activity and characterize different disease stages in multiple sclerosis Ann Neurol 45:695–703
6. Segal BM, Klinman DM, Shevach EM 1997 Microbial products induce autoimmune disease by an IL-12-dependent pathway. J Immunol 158:5087–5090
7. Buyon JP 1998 The effects of pregnancy on autoimmune diseases. J Leukocyte Biol 63:281–287
8. Confavreux C, Hutchinson M, Hours MM, Cortinovis-Tourniaire P, Moreau T 1998 Rate of pregnancy-related relapse in multiple sclerosis. Pregnancy in Multiple Sclerosis Group. N Engl J Med 339:285–291
9. Wilder RL 1995 Neuroendocrine-immune system interactions and autoimmunity. Annu Rev Immunol 13:307–338
10. Silman A, Kay A, Brennan P 1992 Timing of pregnancy in relation to the onset of rheumatoid arthritis. Arthritis Rheum 35:152–155
11. Chrousos GP, Torpy DJ, Gold PW 1998 Interactions between the hypothalamic-pituitary-adrenal axis and the female reproductive system: clinical implications. Ann Intern Med 129:229–240
12. Elenkov IJ, Papanicolaou DA, Wilder RL, Chrousos GP 1996 Modulatory effects of glucocorticoids and catecholamines on human interleukin-12 and interleukin-10 production: clinical implications. Proc Assoc Am Physicians 108:374–381
13. Bieth MH, DeKruyff RH, Umetsu DT 1997 Corticosteroids inhibit IL-12 production in human monocytes and enhance their capacity to induce IL-4 synthesis in CD4+ lymphocytes. J Immunol 158:5589–5595
14. Panina-Bordignon P, Mazzeo D, Lucia PD, et al. 1997 Beta2-agonists prevent Th1 development by selective inhibition of interleukin 12. J Clin Invest 100:1513–1519
15. Lemire JM, Archer DC, Beck L, Spiegelberg HL 1995 Immunosuppressive actions of 1,25-dihydroxyvitamin D3: preferential inhibition of Th1 functions. J Nutr 125:1704S–1708S
16. D'Ambrosio D, Cippitelli M, Cocciolo MG, et al. 1998. Inhibition of IL-12 production by 1,25-dihydroxyvitamin D3: involvement of NF-kappaB downregulation in transcriptional repression of the p40 gene. J Clin Invest 101:252–262
17. Elenkov IJ, Hoffman J, Wilder RL 1997 Does differential neuroendocrine control of cytokine production govern the expression of autoimmune diseases in pregnancy and the postpartum period? Mol Med Today 3:379–383.
18. Westendorp RG, Langermans JA, Huizinga TW, et al. 1997 Genetic influences on cytokine production and fatal meningococcal disease. Lancet 349:170–173
19. Entzian P, Linnemann K, Schlaak M, Zabel P 1996 Obstructive sleep apnea syndrome and circadian rhythms of hormones and cytokines. Am J Respir Crit Care Med 153:1080–1086
20. Chehimi J, Starr SE, Frank I, et al. 1994. Impaired interleukin 12 production in human immunodeficiency virus-infected patients. J Exp Med 179:1361–1366
21. Jacob CO, Fronek Z, Lewis GD, Koo M, Hansen JA, McDevitt HO 1990 Heritable major histocompatibility complex class II-associated differences in production of tumor necrosis factor alpha: relevance to genetic predisposition to systemic lupus erythematosus. Proc Natl Acad Sci USA 87:1233–1237
22. Elenkov IJ, Webster E, Papanicolaou DA, Fleisher TA, Chrousos GP, Wilder RL 1998 Histamine potently suppresses human IL-12 and stimulates IL-10 production via H2 receptors. J Immunol 161:2586–2593
23. Russell AS, Johnston C, Chew C, Maksymowych WP 1997 Evidence for reduced Th1 function in normal pregnancy: a hypothesis for the remission of rheumatoid arthritis. J Rheumatol 24:1045–1050
24. Marzi M, Vigano A, Trabattoni D, et al. 1996 Characterization of type 1 and type 2 cytokine production profile in physiologic and pathologic human pregnancy. Clin Exp Immunol 106:127–133
25. Cadet P, Rady PL, Tyring SK, Yandell RB, Hughes TK 1995 Interleukin-10 messenger ribonucleic acid in human placenta: implications of a role for interleukin-10 in fetal allograft protection. Am J Obstet Gynecol 173:25–29
26. Cohen WR, Galen LH, Vega-Eich M, Young JB 1988 Cardiac sympathetic activity during rat pregnancy. Metabolism 37:771–777
27. Halhali A, Diaz L, Sanchez I, Garabedian M, Bourges H, Larrea F 1999 Effects of IGF-I on 1,25-dihydroxyvitamin D(3) synthesis by human placenta in culture. Mol Hum Reprod 5:771–776
28. Seely EW, Brown EM, DeMaggio DM, Weldon DK, Graves SW 1997 A prospective study of calciotropic hormones in pregnancy and post partum: reciprocal changes in serum intact parathyroid hormone and 1,25-dihydroxyvitamin D. Am J Obstet Gynecol 176:214–217
29. Elenkov IJ, Chrousos GP 1999 Stress hormones, Th1/Th2 patterns, pro/anti-inflammatory cytokines and susceptibility to disease. Trends Endocrinol Metab 10:359–368
30. Ramierz F, Fowell DJ, Puklavec M, Simmonds S, Mason D 1996 Glucocorticoids promote a TH2 cytokine response by CD4+ T cells in vitro. J Immunol 156:2406–2412
31. Salt PJ 1972 Inhibition of noradrenaline uptake 2 in the isolated rat heart by steroids, clonidine and methoxylated phenylethylamines. Eur J Pharmacol 20:329–340
32. Piccinni MP, Giudizi MG, Biagiotti R, et al. 1995 Progesterone favors the development of human T helper cells producing Th2-type cytokines and promotes both IL-4 production and membrane CD30 expression in established Th1 cell clones. J Immunol 155:128–133
33. Gilmore W, Weiner LP, Correale J 1997 Effect of estradiol on cytokine secretion by proteolipid protein-specific T cell clones isolated from multiple sclerosis patients and normal control subjects. J Immunol 158:446–451

Review Article

Hormonal Changes in the Postpartum and Implications for Postpartum Depression

VICTORIA HENDRICK, M.D.
LORI L. ALTSHULER, M.D.
RITA SURI, M.D.

The months following childbirth are a time of heightened vulnerability to depressive mood changes. Because of the abrupt and dramatic changes occurring in hormone levels after delivery, many studies have examined the role of hormonal factors in postpartum depression. The authors review the literature on potential hormonal etiologies in postpartum depression, in particular for progesterone, estrogen, prolactin, cortisol, oxytocin, thyroid, and vasopressin. While evidence for an etiologic role is lacking for most hormones, changes in certain hormonal axes may contribute to depressive mood changes in some women following childbirth. (Psychosomatics 1998; 39:93–101)

The weeks following childbirth are a time of vulnerability to depressive symptomatology in women.[1,2] The literature on postpartum depression has inconsistently defined its time of onset from between 4 weeks and 6 months following delivery. DSM-IV, in an attempt to define the syndrome more rigorously, applies the term "postpartum onset" to depression occurring within 4 weeks of delivery. Most epidemiologic studies have not used this strict criterion. When defined as depression occurring in the first 6 months after delivery, rates are as high as 22%,[3] but drop to 12% to 16% if defined more narrowly as occurring in the first 6 to 9 weeks postpartum.[3,4]

Aside from the postpartum specifier, DSM-IV's criteria for postpartum depression are no different from those of a major depressive episode. However, in comparison with depression occurring at other times in women's lives, guilt and agitation appear to occur more frequently in cases of postpartum depression, and suicidality is less common.[5]

Risk factors for postpartum depression include a family history and a personal history of major depression[6] and depressive symptomatology during pregnancy.[7] Marital discord and stressful child care events (e.g., health problems in the baby) also increase the likelihood of postpartum depression.[6,7] A number of studies have explored whether specific biological characteristics may underlie depression in the postpartum, but with equivocal results. This article reviews the literature on hormonal factors that have been postulated as etiologic in postpartum depression.

HORMONAL EVENTS IN PREGNANCY AND POSTPARTUM

During pregnancy, levels of estrogens (estradiol, estriol, and estrone) and progesterone rise stead-

Received January 24, 1997; revised May 25, 1997; accepted June 6, 1997. From the University of California, Los Angeles (UCLA), Neuropsychiatric Institute and Hospital, and West Los Angeles Veterans Affairs Medical Center, Los Angeles, California. Address reprint requests to Dr. Hendrick, UCLA Neuropsychiatric Institute and Hospital, 300 Medical Plaza, Suite 1544, Los Angeles, CA 90095–6968.

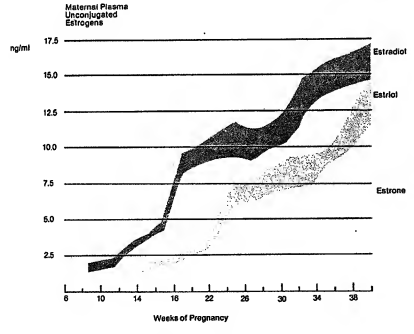
ily (see Figure 1 and Figure 2),[8] in large part as a result of placental production of these hormones. With removal of the placenta at delivery, estrogen and progesterone levels drop sharply, reaching pregravid levels by the fifth postpartum day. Levels of beta-endorphin, human chorionic gonadotrophin, and cortisol also rise across pregnancy, reaching a maximum near term and declining at delivery.

High estrogen levels during pregnancy stimulate production of thyroid hormone-binding globulin, leading to a rise in levels of bound T_3 (triiodothyronine) and T_4 (thyroxine) and a simultaneous drop in levels of free T_3 and T_4. In consequence, thyroid-stimulating hormone (TSH) increases to compensate for the low free-thyroid hormones, and free T_3 and T_4 thus remain within the normal range.[9] With the drop in thyroid-binding globulin following delivery, levels of total T_3 and T_4 drop, whereas free T_3 and T_4 remain relatively constant. Prolactin levels rise during pregnancy, peak at delivery and, in nonlactating women, return to pregravid levels within 3 weeks postpartum. By inducing the release of oxytocin, a hormone that stimulates pituitary lactotrophic cells, breast-feeding maintains high prolactin levels. Even in breast-feeding women, however, prolactin levels eventually return to pregravid levels.

GONADAL STEROIDS

Estradiol and estriol are biologically active forms of estrogen that are produced by the placenta and rise during pregnancy by 100-fold and 1,000-fold, respectively. Because synthesis of estriol results from metabolic activity of the fetal liver, it is produced in high concentrations during pregnancy. Animal studies have demonstrated that estradiol enhances neurotransmitter

FIGURE 1. Rise in levels of estrogens during pregnancy



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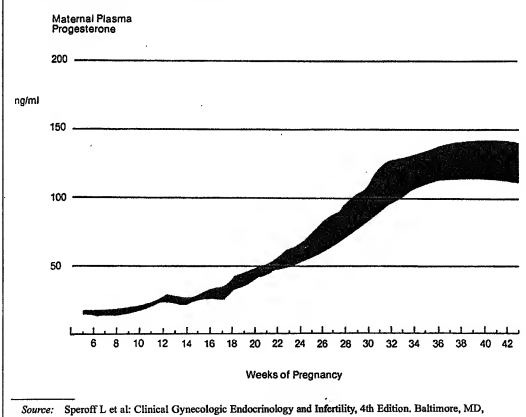
function through increased synthesis and reduced breakdown of serotonin.[10] The abrupt decrease in estradiol levels following delivery may thus theoretically contribute to postpartum depression. However, a study of 182 childbearing women found no significant difference in the magnitude of change of total estradiol or of free estriol from late pregnancy to the puerperium in depressed and nondepressed women.[11] Total estradiol levels, measured on 9 separate days from Week 34 of gestation to Postpartum Day 8, were no different among the 2 groups of women, with the exception of a single significantly lower level of total estradiol at Week 36 in the women who developed postpartum depression. This finding is of unclear significance, particularly as the lower level was found in an antepartum rather than a postpartum sample. Other studies of total estradiol levels, obtained at various times between the first day and the eighth week following delivery, have found no difference in women with and without postpartum depression.[12,13] Levels of unbound (free) estradiol have not been studied in women with postpartum depression but merit examination, as the unbound form is biologically active.

Two recent studies have reported that estrogen supplementation significantly reduced postpartum depressive symptoms. The first was a small open study that included four women with a history of postpartum depression.[14] In the month following delivery the women received up to 10 mg of Premarin (estrogen-replacement therapy) daily, equivalent to about 15 times the

FIGURE 2. Rise in level of progesterone during pregnancy



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usual dose for estrogen-deficiency symptoms, and thus required heparin (5,000 units bid) to prevent thromboembolic phenomena. Over a 12-month follow-up period, none of these women experienced a recurrence of postpartum depression, despite the expected risk of relapse of 35% to 60%. The small sample size (4 cases of postpartum depression) was a major limitation of the study. In the second, a double-blind placebo-controlled study of 61 women with major depression that developed within 3 months of delivery,[15] 80% of the patients receiving an estrogen patch had Edinburgh Postnatal Depression Scale scores under the threshold for major depression after 3 months of treatment, compared with 31% of the placebo-treated group. However, nearly half of the estrogen-treated patients were also on antidepressant medications, confounding the study results.

The sharp decline in progesterone levels following childbirth has also been implicated in postpartum mood changes, but the data are conflicting. A study of 27 women followed every 3 days for the first 6 weeks after delivery found a weak association between postpartum depression and the magnitude of change of progesterone.[12] Further studies, however, have failed to confirm a relationship between postpartum depression and blood levels of either total[16,17] or free progesterone.[11,18]

Salivary levels of progesterone have been examined on the premise that they reflect the free, biologically active, fraction of plasma progesterone concentrations. A study of 147 mothers at 6 to 8 weeks postpartum found that the depressed breast-feeding women had lower levels of salivary progesterone than the euthymic breast-feeding women.[13] Levels of salivary progesterone were higher, on the other hand, in depressed postpartum women who were bottle feeding. However, nursing may have influenced progesterone levels by suppressing menstrual cycling, confounding the results of the study. A prospective study of 120 women found no association between the levels or the magnitude of change of salivary progesterone and depression at Day 35 postpartum.[19] One report describes prophylactic efficacy of progesterone given postnatally, but this study lacked a control group.[20] No controlled studies exist to date of progesterone in the prophylaxis or treatment of postpartum depression.

THYROID HORMONES

The incidence of abnormal thyroid function rises slightly after childbirth. In the 6 months following delivery, women experience thyroid dysfunction at a rate of up to 7%,[21–23] compared with a rate of 3% to 4% in the general population.[24] Although thyroid dysfunction has not been identified in most women with postpartum depression,[25] it may play a role for a subgroup of women.[26–31] In a prospective study of 303 pregnant euthyroid women, 21 women (7%) developed postpartum thyroid disorders.[31] Depression was identified in 38% of these 21 mothers and resolved with treatment of the thyroid dysfunction.[31] Thus, in women with symptoms suggesting hypothyroidism (weight gain, cold intolerance, lethargy), measurement of thyroid function is an important part of the evaluation of postpartum depression.

Some postpartum women without overt thyroid dysfunction may nevertheless have thyroid pathology. Thyroid antibodies have been found in up to 11.6% of postpartum women.[27] The immunosuppressant effect of high cortisol levels during pregnancy may be followed by a "rebound" immune phenomenon after delivery, producing a high incidence of postpartum thyroid antibodies.[9] A double-blind study of 145 antibody-positive women and 229 antibody-negative women found a relationship between depression and postpartum antibody status.[27] At 6 weeks following delivery, 43% of the antibody-positive women had mild-to-moderate depressive symptoms, compared with 28% of the antibody-negative women. Depression was defined by a score of 17 or higher on the Hamilton Depression scale, a score of 13 or more on the Edinburgh postnatal depression scale, and a score of 11 or more on a hospital anxiety and depression scale. Antibody-positive women should be followed with thyroid function testing beyond the postpartum period, as many patients

with antithyroid antibodies go on to develop overt hypothyroidism within 4 years.[32] At this time, however, there does not appear to be a role for thyroid antibody testing in the postpartum, as the relationship between antibodies and depression is weak.

Diminished thyroid function may affect postpartum mood through its association with diminished central 5-HT (5-hydroxytryptamine [serotonin]) activity. Blood levels of 5-HT have been positively correlated with thyroid hormone levels,[33] and the prolactin and cortisol responses to the 5-HT agonist fenfluramine are blunted in hypothyroid patients compared with euthyroid controls, suggesting reduced central 5-HT activity.[34]

PITUITARY HORMONES

Prolactin rises from pregravid levels of 5–25 ng/ml to 140 ng/ml in late pregnancy and drops in the 3 weeks after delivery in nonlactating women. In breast-feeding mothers, prolactin levels remain high for several months but eventually decline to prepregnancy levels. Prolactin's role in psychopathology has been suggested by the association of anxiety, depression, and hostility in nonpregnant women with pathologic hyperprolactinemia compared with control subjects.[35] One study of 147 women at 6–8 weeks postpartum found lower prolactin levels in the depressed breast-feeding women than in the nondepressed breast-feeding women.[13] However, all levels remained within normal physiological ranges. The study did not control for the relationship between breast-feeding and sampling time. As prolactin levels increase following breast-feeding and nipple stimulation, this is a significant confound. A large prospective study that did control for breast-feeding, in addition to demographic and psychosocial variables, failed to find a relationship between prolactin levels and postpartum mood.[11]

Oxytocin and vasopressin, two posterior pituitary hormones that undergo changes in levels in the postpartum, have not been assessed for their relationship to postpartum depression. Oxytocin, which rises sharply at delivery and with breast-feeding, stimulates uterine muscle contraction at labor and promotes release of breast milk. In animal studies, oxytocin also appears to stimulate maternal behavior.

Vasopressin regulates blood pressure and electrolyte balance and has been found lower in urine, but not plasma, of postpartum women compared with the nonpuerperal women in a study that did not assess mood state.[36] While negative results have been found in studies of vasopressin levels in women with postpartum blues,[36,37] no studies have assessed its levels in postpartum depression.

CORTISOL

Cortisol levels peak in late pregnancy as a result of placental production of corticotropin releasing hormone, and fall abruptly at delivery. A number of studies have failed to find an association between plasma cortisol,[11,13,38,39] or urinary-free cortisol[11] and postpartum depression. One study that did note a positive association between morning serum cortisol levels at 6 weeks postpartum and degree of dysphoria in 26 women[29] was confounded by a lack of control for stressful life events and for timing of breast-feeding, factors that may produce an elevation or a reduction, respectively, of cortisol levels.[40]

A prospective study of 182 women followed from the second trimester of pregnancy until Postpartum Week 9 controlled for lactation and for demographic, psychiatric, social, life stress, and other variables. No association was observed between total cortisol, urinary-free cortisol, or dexamethasone-suppression test results and postpartum mood.[11] Thus, current data do not support an etiologic role for cortisol in the onset of postpartum depression. A prospective study of 17 healty euthymic women evaluated in the second trimester of pregnancy and followed to the 12th postpartum week similarly found no relationship between mood and cortisol levels but did observe a significantly greater and longer lasting blunting of adrenocorticotropic hormone (ACTH) response to corticotropin-releasing hormone in women who developed postpartum blues or postpartum depression

compared with women who remained euthymic.[41] The authors speculated that the hypercortisolism that characterizes late pregnancy (resulting from placental production of corticotropin-releasing hormone) produces adrenal suppression following delivery that, when sustained and severe, may contribute to depressive mood changes after delivery. This intriguing but small study, consisting of only one case of postpartum depression and seven cases of postpartum blues, merits examination with a larger sample size.

DISCUSSION AND CONCLUSIONS

The dramatic physiological events occurring after delivery have led researchers to speculate that postpartum mood disorders result from a biochemical or hormonal etiology. While certain hormones, such as estradiol and ACTH, merit further exploration, studies have been negative or contradictory for most biological variables thought to be etiologic. Thus, the literature to date does not consistently support any single biological etiology for postpartum depression.

Methodological problems in many studies may have led to the conflicting results. For example, blood sampling in many studies did not control for breast-feeding. Lactation not only influences levels of prolactin, progesterone, estrogen, oxytocin, and cortisol but also has been associated with changes in mood state, both positive[42] and negative.[43] Other variables seldom controlled for in the studies were the time of day when assays were obtained, seasonal variations in hormone levels, extent of sleep deprivation in the mother, and potential medication effects on hormone levels. Many studies assessed total hormone concentration rather than free, biologically active hormone levels. While the majority of studies measured the absolute levels of a biological factor, it may be the degree of change—in particular, the degree of change of free hormone—from pregnancy to the early postpartum that affects psychopathology. Changes in mood may also occur from extreme sensitivity to normal levels of hormones.

A limitation of studies assessing serum levels of hormones and other biological factors is that peripheral levels do not necessarily reflect central activity. For beta-endorphins, for example, the relationship between peripheral and cerebrospinal fluid concentrations is small.[44] Thyroid hormone measures similarly show little parallel with peripheral indices.[45,46] Thus, measurement techniques that reliably reflect central neurotransmission are necessary to better establish the relationship between postpartum mood changes and neurotransmitter activity. Central levels of steroid hormones, however, are reported to correlate with plasma levels.[47]

It is possible that no biological etiologies are specific to the postpartum, but rather the birth of a child may represent a major stressful life event that, in vulnerable women, precipitates a depressive episode. Clearly, psychosocial stressors contribute to the syndrome in many women: a lack of support, marital conflict, unemployment, an unplanned pregnancy, single motherhood, and younger age are some factors associated with postpartum depression.[2,42,48] Infant factors, including high levels of irritability and poor motor behavior, also increase the likelihood of maternal depression.[49,50] Future research on the biological factors that may underlie postpartum mood disorders should attempt to control for these variables, as they otherwise are likely to confound the data. Measures such as the Neonatal Behavioral Assessment Scale,[51] the Life Events and Difficulties Schedule,[52] and the Perceived Stress Scale[53] can be used for this purpose. Further variables that should be taken into account include personality traits in the mother, length of time and severity of the mother's depression, and qualititative aspects of the depression, including presence of obsessional or anxious features. These are factors that have been shown to predict likelihood of antidepressant response in nonpuerperal major depression but their role in influencing treatment outcome of postpartum depression has not been assessed. The genetic vulnerability that may underlie the development of depression in the postpartum is also worth investigating, for example, through the use of family studies of women with postpartum depression.

A significant problem in research on the etiology of postpartum depression is the heterogeneity of the syndrome. Depression arising 1 week postpartum may be etiologically different from depression developing 3 months after delivery or from depression that had its onset during the pregnancy but continued through the postpartum period. Further, a postpartum depression with anxious and obsessional features may be etiologically different from an anergic postpartum depression. Some authors have postulated that postpartum depressions exist in two distinct categories: cases in which the index episode occurs in the postpartum, and cases in which the postpartum depression represents a recurrence of a previous nonpuerperal depression.[54] Compared with the former, the latter group appears to have a greater likelihood of nonpuerperal recurrence[54,55] and thus may require closer long-term follow-up. Other treatment implications, such as differences in treatment outcome between the two groups, are not clear. To our knowledge, no studies have examined biological variables that may distinguish these two potentially distinct populations of postpartum depressed women.

Postpartum depression can produce significant distress to the new mother and her family and may have an adverse impact on the cognitive and emotional development of the child.[56,57] Further, a postpartum depression predisposes a woman to future psychopathology, particularly following subsequent deliveries. The identification of etiologic factors is therefore of great importance, to allow for better understanding of preventive and treatment strategies. With the increasing tendency for researchers to employ standardized rating instruments (Edinburgh Postnatal Depression Scale, etc.) and to adhere to stringent criteria for postpartum depression, research in the etiology of postpartum depression is likely to advance in coming years.

References

1. Stowe ZN, Nemeroff CB: Women at risk for postpartum-onset major depression. Am J Obstet Gynecol 1995; 173:639–645
2. O'Hara MW: Postpartum depression: causes and consequences. New York, Springer-Verlag, 1995
3. Watson JP, Elliott SA, Rugg AJ, et al: Psychiatric disorder in pregnancy and the first postnatal year. Br J Psychiatry 1984; 144:453–462
4. O'Hara MW: Social support, life events, and depression during pregnancy and the puerperium. Arch Gen Psychiatry 1986; 43:569–573
5. Whiffen VE: Is postpartum depression a distinct diagnosis? Clinical Psychology Review 1992; 12:485–508
6. O'Hara MW, Neunaber DJ, Zekoski EM: Prospective study of postpartum depression: prevalence, course, and predictive factors. J Abnorm Psychol 1984; 93:158–171
7. Gotlib IH, Whiffen VE, Wallace PM, et al: Prospective investigation of postpartum depression: factors involved in onset and recovery. J Abnorm Psychol 1991; 100:122–132
8. Speroff L, Glass RH, Kase NG: Clinical Gynecologic Endocrinology and Infertility, 3rd Edition. Baltimore, MD, Williams & Wilkins., 1983
9. Learoyd DL, Fung HYM, McGregor AM: Postpartum thyroid dysfunction. Thyroid 1992; 2:73–80
10. Luine VN, Khylchevskaya RI, McEven B: Effect of gonadal steroids on activities of monoamine oxidase and choline acetylase in rat brain. Brain Res 1975; 86:293–306
11. O'Hara MW, Schlechte JA, Lewis DA, et al: Controlled prospective study of postpartum mood disorders: psychological, environmental, and hormonal variables. J Abnorm Psychol 1991; 100:63–73
12. Nott PN, Franklin M, Armitage C, et al: Hormonal changes and mood in the puerperium. Br J Psychiatry 1976; 128:379–383
13. Harris B, Johns S, Fung H, et al: The hormonal environment of postnatal depression. Br J Psychiatry 1989; 154:660–667
14. Sichel DA, Cohen LS, Robertson LM, et al: Prophylactic estrogen in recurrent postpartum affective disorder. Biol Psychiatry 1995; 38:814–818
15. Gregoire AJP, Kuman R, Everitt B, et al: Transdermal oestrogen for treatment of severe postnatal depression. Lancet 1996; 347:930–933
16. Butler J, Leonard BE: Postpartum depression and the effect of nomifensine treatment. Int Clin Psychopharmacol 1986; 1:244–252
17. Ballinger CB, Kay DSG, Naylor GJ, et al: Some biochemical findings during pregnancy and after delivery in relation to mood change. Psychol Med 1982; 12:549–556
18. Heidrich A, Schleyer M, Spingler H, et al: Postpartum blues: relationship between not-protein bound steroid

hormones in plasma and postpartum mood changes. J Affect Disord 1994; 30:93–98
19. Harris B, Lovett L, Newcombe R, et al: Maternity blues and major endocrine changes: Cardiff puerperal mood and hormone study. BMJ 1994; 308:949–953
20. Dalton K: Progesterone prophylaxis used successfully in postnatal depression. Practitioner 1985; 229:507–508
21. Pop VJM, Derooy HAM, Bader HL, et al: Microsomal antibodies during gestation in relation to postpartum thyroid dysfunction and depression. Acta Endocrinologica 1993; 129:26–30
22. Goldman JM: Postpartum thyroid dysfunction. Arch Intern Med 1986; 146:1296–1299
23. Amino N, Mori H, Iwatani Y, et al: High prevalence of transient postpartum thyrotoxicosis and hypothyroidism. N Engl J Med 1982; 306:849–852
24. Hershman JM: Thyroid disease, in Endocrine Pathophysiology, edited by Hershman JM. Philadelphia, PA, Lea & Febiger, 1988, pp. 37–76
25. George AJ, Wilson KCM: Beta-endorphin and puerperal psychiatric symptoms. Br J Pharmacol 1983; 80:493
26. Harris B, Fung H, Johns S, et al: Transient postpartum thyroid dysfunction and thyroid antibodies and depression. J Affect Disord 1989; 17:243–249
27. Harris B, Othman S, Davies JA, et al: Association between postpartum thyroid dysfunction and thyroid antibodies and depression. BMJ 1992; 305:152–156
28. Harris B, Lovett L, Roberts S, et al: Cardiff puerperal mood and hormone study, paper I: Saliva steroid hormone profiles in late pregnancy and the puerperium: endocrine factors and parturition. Hormone Research 1993; 39:138–145
29. Pedersen CA, Stern RA, Pate J, et al: Thyroid and adrenal measures during late pregnancy and the puerperium in women who have been major depressed or who become dysphoric postpartum. J Affect Disord 1993; 29:201–211
30. Glover V: Do biochemical factors play a part in postnatal depression? Prog Neuropsychopharmacol Biol Psychiatry 1992; 16:605–615
31. Pop VJM, de Rooy HAM, Vader HL, et al: Postpartum thyroid dysfunction and depression in an unselected population. N Engl J Med 1991; 324:1815–1816
32. Surks MI, Ocampo E: Subclinical thyroid disease. Am J Med 1996; 100:217–223
33. Upadhyaha L, Agrawal JK, Dubey GP, et al: Biogenic amines and thyrotoxicosis. Acta Endocrinologia 1992: 126:315–318
34. Cleare AJ, McGregor A, O'Kean V: Neuroendocrine evidence for an association between hypothyroidism, reduced central 5HT activity, and depression. Clin Endocrinol (Oxf) 1995; 43:713–719
35. Fava M, Fava GA, Kellner R, et al: Psychosomatic aspects of hyperprolactinemia. Psychother Psychosom 1983; 40:257–262
36. Stein G, Morton J, Marsh A, et al: Vasopressin and mood during the puerperium. Biol Psychiatry 1984; 19:1711–1717
37. Stein G, Marsh A, Morton J: Mental symptoms, weight changes, and electrolyte excretion in the first postpartum week. J Psychosom Res 1981; 25:395–408
38. Gard PR, Handley SL, Parsons AD, et al: A multivariate investigation of postpartum mood disturbance. Br Journal Psychiatry 1986; 148:567–575
39. Handley SL, Dunn TL, Waldron G, et al: Tryptophan, cortisol and puerperal mood. Br J Psychiatry 1980; 136:498–508
40. Altemus M, Deuster PA, Galliven E, et al: Suppression of hypothalamic-pituitary-adrenal axis responses to stress in lactating women. J Clin Endocrinol Metab 1995; 80:2954–2959
41. Magiakou MA, Mastorakos G, Rabin D, et al: Hypothalamic corticotropin-releasing hormone suppression during the postpartum period: implications for the increase in psychiatric manifestations at this time. J Clin Endocrinol Metab 1996; 81:1912–1917
42. Warner R, Appleby L, Whitton A, et al: Demographic and obstetric risk factors for postnatal psychiatric morbidity. Br J Psychiatry 1996; 168:607–611
43. Alder EM, Cox JL: Breast feeding and postnatal depression. J Psychosom Res 1983; 27:139–144
44. Smith R, Owens PC, Lovelock M, et al: Acute hemorrhagic stress in conscious sheep elevates immunoreactive beta-endorphin in plasma but not in cerebrospinal fluid. Endocrinology 1986; 118:2572–2576
45. Andersson K, Eneroth P: The effects of acute and chronic treatment with triiodothyronine and thyroxine on the hypothalamic and telecncephalic catecholamine nerve terminal systems of the hypophysectomized male rat. Neuroendocrinology 1985; 40:398–408
46. Barsano C, Garces J, Iqbal Z: Metabolic implications of low dose triiodothyronine administration in rats: relevance to the adjunctive use of triiodothyronine in the treatment of depression. Biol Psychiatry 1994; 35:814–823
47. Backstrom T, Carstensen H, Sodergard R: Concentration of estradiol, testosterone and progesterone in cerebrospinal fluid compared to plasma unbound and total concentrations. J Steroid Biochem 1976; 7:469–472
48. Gotlib IH, Whiffen VE, Mount JH, et al: Prevalence rates and demographic characteristics associated with depression in pregnancy and the postpartum. J Consult Clin Psychol 1989; 57:269–274
49. Murray L, Stanley C, Hooper R, et al: The role of infant factors in postnatal depression and mother-infant interactions. Dev Med Child Neurol 1996; 38:109–119
50. Murray L, Cooper PJ: The role of infant and maternal factors in postpartum depression, mother-infant interactions, and infant outcome, in Postpartum Depression and Child Development, edited by Murray L, Cooper P. New York, Guilford, 1997, pp. 111–135
51. Brazelton TB: Neonatal Behavioral Assessment Scale,

2nd Edition. Clinics in Developmental Medicine, No. 88. London, England, S.I.M.P., 1984
52. Brown GW, Harris T: Social Origins of Depression: A Study of Psychiatric Disorder in Women. London, England, Tavistock, 1978
53. Cohen S, Kamarck T, Mermelstein R: A global measure of perceived stress. J Health Soc Behav 1983; 24:385–396
54. Cooper PJ, Murray L:Course and recurrence of postnatal depression. Evidence for the specificity of a diagnostic concept. Br J Psychiatry 1995; 166:191–195
55. Bell AJ, Land NM, Milner S,et al: Long-term outcome of postpartum psychiatric illness requiring admission. J Affect Disord 1994; 31:67–70
56. Field T: Infants of depressed mothers. Infant Behavior and Development 1995; 18:1–13
57. Coghill SR, Caplan HL, Alexandra H, et al: Impact of maternal postnatal depression on cognitive development of young children. BMJ 1986; 292:1165–1167

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[N] [O] [P] [Q] [R] [S] [T] [U] [V] [W] [X] [Y] [Z]

The pdf online version of the IUPAC Compendium of Chemical Terminology mostly corresponds to the second edition (1997), compiled by Alan D. McNaught and Andrew Wilkinson (Royal Society of Chemistry, Cambridge, UK). Towards the end of 2003, work began on the addition of terms from more recent IUPAC recommendations, with the intention of eventually bringing the website into a condition in which it can be maintained up-to-date.

Some minor errors have been corrected (the changes are noted where they occur), and cross-referencing has been improved. The conversion to electronic form (pdf files) was carried out by David Stout (Information Technology Consultant, Information Services, Royal Society of Chemistry).

Exhibit H
2 Pages Total

moiety
In physical organic chemistry moiety is generally used to signify part of a molecule, e.g. in an ester R^1COOR^2 the alcohol moiety is R^2O. The term should not be used for a small fragment of a molecule.
1994, *66*, 1141

INTERNATIONAL UNION OF
PURE AND APPLIED CHEMISTRY
AND
INTERNATIONAL UNION OF BIOCHEMISTRY

DEFINITIVE RULES FOR NOMENCLATURE OF STEROIDS

Issued by the
IUPAC Commission on the Nomenclature of Organic Chemistry

and

IUPAC–IUB Commission on Biochemical Nomenclature

1971

LONDON
BUTTERWORTHS

P.A.C.—31—1/2—M

Exhibit I
5 Pages total

DEFINITIVE RULES FOR NOMENCLATURE OF STEROIDS†

CONTENTS

INTRODUCTION

The rules for steroid nomenclature originate from a discussion held at the Ciba Foundation in London, England, in 1950 between the representatives of many schools. These were published in *Chem. & Ind., London*, (1951) pp SN 1–11, and also in French and German. They were subsequently taken over by the International Union of Pure and Applied Chemistry and published in an official form in the *Comptes Rendus* of the Zürich meeting in 1952

† These Rules shall be known as the IUPAC–IUB 1971 Definitive Rules for Steroid Nomenclature.

These Rules are issued by the IUPAC Commission on the Nomenclature of Organic Chemistry and by the IUPAC–IUB Commission on Biochemical Nomenclature.

Those who have served on the Commission on the Nomenclature of Organic Chemistry for varying periods during 1967–71 are the following. Present members are shown by an asterisk*. P. E. Verkade (Chairman to 1971), N. Lozac'h* (Chairman from 1971), K. Bláha*, L. C. Cross*, G. M. Dyson, S. P. Klesney*, W. Klyne*, K. L. Loening*, H. S. Nutting, J. Rigaudy*, S. Veibel*. Associate members: R. S. Cahn, H. Grünewald*, K. Hirayama*. Observer: K. A. Jensen*.

Those who have served on the Commission on Biochemical Nomenclature for varying periods during 1967–71 are the following. Present members are shown by an asterisk*. O. Hoffmann-Ostenhof* (Chairman), A. E. Braunstein*, W. E. Cohn*, J. S. Fruton, B. Horecker*, P. Karlson*, B. Keil*, W. Klyne*, C. Liébecq*, E. C. Slater, E. C. Webb*, W. J. Whelan*. Observer: S. Veibel*.

Comments on and suggestions for future revisions of these Rules should be sent to: Professor N. Lozac'h, Ecole nationale supérieure de Chimie, 5 Avenue d'Edimbourg, F-14 Caen, France, or Professor O. Hoffmann-Ostenhof, Lehrkanzel für Biochemie der Universität Wien, Währingerstrasse 38, 1090 Vienna, Austria, or to any present member of the Commissions named above.

[also *IUPAC* 1957 *Rules for Nomenclature of Steroids*, Butterworths: London (1958); 2nd ed. 1966, pp 71–82; and numerous reprints and translations, including *J. Am. Chem. Soc.* **82**, 5577 (1960)].

In 1960 a group of specialists under the chairmanship of Professor T. Reichstein, including representatives of the IUPAC Commissions on the Nomenclature of Organic Chemistry and of Biochemical Nomenclature, met in Basle, Switzerland, for discussions of amendments and additions to the Rules. Agreement was not reached on all the points discussed, and the results of this meeting were therefore published in discussion form in the *IUPAC Information Bulletin*, No. 11. They have generally been referred to as the 'Basle Proposals'.

Since then, many points in the Basle Proposals have become almost universally accepted in the literature. In 1965 the two International Commissions concerned, namely, the IUPAC Commission on the Nomenclature of Organic Chemistry and the Commission on Biochemical Nomenclature (now jointly responsible to IUPAC and IUB), decided that the time had come for as many as possible of the Basle Proposals to be formulated as rules. Accordingly Tentative Rules were formulated and published in *Biochim. Biophys. Acta*, **164**, 453–486 (1968), in *IUPAC Information Bulletin*, No. 33 (1968), and elsewhere. These Rules have subsequently been studied by the two Commissions and amended on a number of (mostly minor) points.

The Definitive Rules include: all the original Rules, mostly renumbered (with additions and amendments arising from the Basle Proposals or from current practice in the literature); and most of the Basle Proposals, namely, those that have been generally accepted. Further, adoption of the sequence-rule procedure* for general stereochemical descriptions in much of the chemical literature has permitted its introduction now also for some sections of steroid nomenclature that were previously in dispute or intractable.

GENERAL APPLICATION

Although these Rules are called 'Rules for Nomenclature of Steroids', many of the principles therein have become universally accepted also in diterpene and triterpene chemistry; also to some extent for sesquiterpenes and for several groups of alkaloids. It is suggested that the same principles may be applied to a number of other specialized groups of natural products, perhaps without the need for further official rules, so long as the basic ideas are followed. These principles include: (i) clear definition of stem names and the stereochemistry implied in them; (ii) systematic application of the rules of general organic chemical nomenclature, with modifications where special considerations make this necessary; (iii) application of the methods of skeletal modification given in these Rules, viz. the use of homo and nor for, respectively, stepwise expansion and contraction of ring systems; the use of seco for reductive fission of ring systems; and the use of *abeo* for formal bond

* R. S. Cahn, C. K. Ingold and V. Prelog, *Angew. Chem. Intern. Ed.* **5**, 385 (1966) (in English); *Angew. Chem.* **78**, 413 (1966) (in German); for a partial simplified account see R. S. Cahn, *J. Chem. Educ.* **41**, 116 (1964). See also 'IUPAC 1968 Tentative Rules for the Nomenclature of Organic Chemistry, Sections E, Fundamental Stereochemistry', *IUPAC Information Bulletin*, No. 35, 71–80 (1969).

migrations (this flexible concept was first proposed by Professor D. H. R. Barton at an informal meeting of terpene chemists convened by the Chemical Society in London, England).

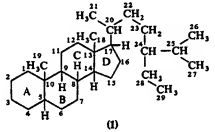
RULES

Rules are numbered **2S–1**, **2S–2**, **2S–3**, etc., the first '**2**' denoting that this is the second or revised set of rules. The numbers of the corresponding previous rules, where they exist, are included for comparison.

GENERAL

Rule 2S–1 (expanded from Rules S–1 and S–2)

1.1. Steroids are numbered and rings are lettered as in Formula (1). If one of the two methyl groups attached to C-25 is substituted it is assigned the lower

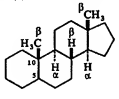


(1)

number (26); if both are substituted, that carrying the substituent cited first in the alphabetical order is assigned the lower number [cf. IUPAC Rule* **C-15.11(e)**]. For trimethyl steroids see Rule **2S–2.3**, Note c.

1.2. If one or more of the carbon atoms shown in (1) is not present and a steroid name is used, the numbering of the remainder is undisturbed.

1.3. For a steroid the name, including stereochemical affixes, and its structural formula (see Rule **2S–1.4**), denote the absolute configuration at each asymmetric centre (see also Rule **2S–1.5**). When the configuration at one or more centres is not known, this is indicated by Greek letter(s) ξ (xi) prefixed by the appropriate numeral(s).

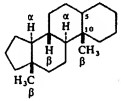


(2)

* *IUPAC Nomenclature of Organic Chemistry, Section A, B and C*, 1971, Butterworths: London.

1.4. When the rings of a steroid are denoted as projections on to the plane of the paper, the formula is normally to be oriented as in (2). An atom or group attached to a ring depicted as in the orientation (2) is termed α (alpha) if it lies below the plane of the paper or β (beta) if it lies above the plane of the paper. In formulae, bonds to atoms or groups lying below the plane of the paper are shown as broken (– – – –) lines, and bonds to atoms or groups lying above the plane of the paper are shown as solid lines preferably thickened (———). Bonds to atoms or groups whose configuration is not known are denoted by wavy lines (~~~~~~~).

Notes: (1) Projections of steroid formulae should not be oriented as in Formula (3), (4) or (5) unless circumstances make it obligatory.



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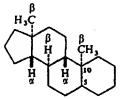

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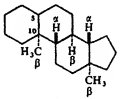
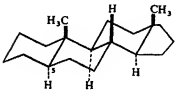


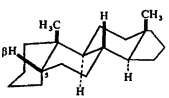
(5)

(2) With the preferred orientation (2), and with (3), α bonds appear as broken lines and β bonds as solid (thickened) lines. The reverse is true for (4) and (5). Wavy lines denote ξ bonds for all orientations of the formula.

(3) A perspective representation of the stereochemistry of Formula (2) as in (2a) or (2b) may also be used.



(2a)
A 5α-steroid



(2b)
A 5β-steroid

(For the significance of the prefixes 5α- and 5β-, see Rule 2S–1.5.)